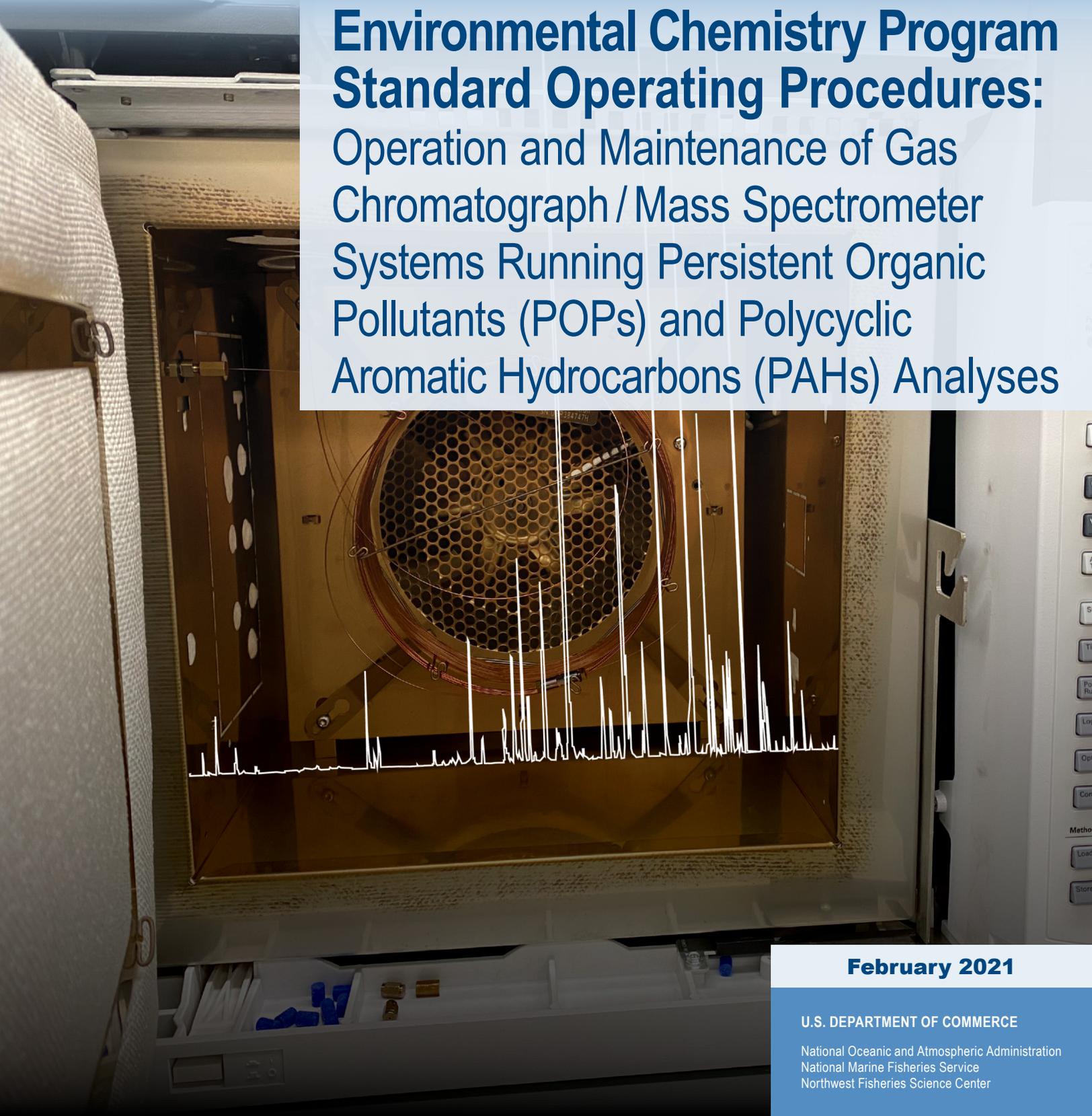




NOAA Processed Report NMFS-NWFSC-PR-2021-01

<https://doi.org/10.25923/fcb1-0r41>

# Environmental Chemistry Program Standard Operating Procedures: Operation and Maintenance of Gas Chromatograph / Mass Spectrometer Systems Running Persistent Organic Pollutants (POPs) and Polycyclic Aromatic Hydrocarbons (PAHs) Analyses



**February 2021**

U.S. DEPARTMENT OF COMMERCE

National Oceanic and Atmospheric Administration  
National Marine Fisheries Service  
Northwest Fisheries Science Center

## NOAA Processed Report Series NMFS-NWFSC-PR

The Northwest Fisheries Science Center of NOAA's National Marine Fisheries Service uses the NOAA Processed Report NMFS-NWFSC-PR series to disseminate information only. Manuscripts have not been peer-reviewed and may be unedited. Documents within this series represent sound professional work, but do not constitute formal publications. They should only be footnoted as a source of information, and may not be cited as formal scientific literature. The data and any conclusions herein are provisional, and may be formally published elsewhere after appropriate review, augmentation, and editing.

NWFSC Processed Reports are available from the NOAA Institutional Repository, <https://repository.library.noaa.gov>.

Mention throughout this document of trade names or commercial companies is for identification purposes only and does not imply endorsement by the National Marine Fisheries Service, NOAA.

Cover image: Agilent 7890A gas chromatography (GC) oven, open, with a chromatogram inside depicting a sample run for POPs analysis. Photograph by J. Gates, NMFS/NWFSC.

### Recommended citation:

(Gates et al. 2021)<sup>1</sup>

<sup>1</sup> Gates, J. B., R. W. Pearce, C. A. Sloan, J. Travis, and I. Schultz. 2021. Environmental Chemistry Program Standard Operating Procedures: Operation and Maintenance of Gas Chromatograph/Mass Spectrometer Systems Running Persistent Organic Pollutants (POPs) and Polycyclic Aromatic Hydrocarbons (PAHs) Analyses. U.S. Department of Commerce, NOAA Processed Report NMFS-NWFSC-PR-2021-01.

<https://doi.org/10.25923/fcb1-0r41>



**NOAA  
FISHERIES**

# **Environmental Chemistry Program Standard Operating Procedures: Operation and Maintenance of Gas Chromatograph / Mass Spectrometer Systems Running Persistent Organic Pollutants (POPs) and Polycyclic Aromatic Hydrocarbons (PAHs) Analyses**

Jonelle B. Gates, Ron W. Pearce, Catherine A. Sloan, Jennifer Travis, and  
Irvin Schultz

<https://doi.org/10.25923/fcb1-0r41>

**February 2021**

Environmental and Fisheries Sciences Division  
Northwest Fisheries Science Center  
2725 Montlake Boulevard East  
Seattle, Washington 98112

**U.S. DEPARTMENT OF COMMERCE**

National Oceanic and Atmospheric Administration  
National Marine Fisheries Service  
Northwest Fisheries Science Center

# Contents

List of Figures .....	iv
List of Printout Examples .....	vi
List of Tables .....	vii
List of Abbreviations and Terminology .....	viii
Acknowledgments .....	ix
1 Introduction .....	1
2 Routine Maintenance .....	2
2.1 Updating the Logbook.....	2
2.2 Cleaning the Inlet.....	3
2.3 Trimming the Guard Column .....	4
2.3.1 Guard Column Maintenance.....	4
2.3.2 Analytical Column Maintenance.....	5
2.3.3 Checking Fittings for Leaks.....	6
2.3.4 Updating Column Length in Methods .....	7
2.4 Changing the Rinse Vials.....	8
2.5 Cleaning the Inlet Inserts .....	8
3 Replacing the Columns .....	9
3.1 Updating the Logbook.....	9
3.2 Replacing the Guard Column .....	9
3.3 Replacing the Analytical Column .....	10
3.3.1 Conditioning a New Analytical Column.....	11
3.3.2 Installing the Analytical Column in the Mass Spectrometer .....	12
3.3.3 Checking the New Column's Column Bleed .....	12
4 Tuning the Mass Spectrometer .....	13
4.1 Updating the Logbook.....	13
4.2 Air / Water Spectrum .....	13
4.2.1 Checking the Air / Water Spectrum Before Tuning.....	13
4.2.2 Checking the Air / Water Spectrum After Tuning.....	14
4.3 Auto Tune ( <b>atune</b> ) and Standard Spectrum Tune ( <b>stune</b> ).....	15
4.4 Standard Spectrum Tune File Used for PAHs Analysis ( <b>sAHtune</b> ) .....	16
4.5 Tune File Used for POPs Analysis ( <b>CHtune</b> ).....	17

5	Running Standards Post-Maintenance .....	18
5.1	Choosing Standards .....	18
5.2	Setting Up and Running the Standards Sequences.....	18
5.3	Checking the Standards.....	19
5.3.1	Standard Used to Check Peak Shape for PAHs Analysis ( <b>AHP4A</b> ) .....	19
5.3.2	Standard Used to Check Sensitivity for PAHs Analysis ( <b>AHP1A</b> ).....	20
5.3.3	Standard Used to Check Peak Shape and Sensitivity for POPs Analysis ( <b>CHBr2</b> ).....	21
5.4	Updating SIM Window Times.....	22
5.4.1	PAHs SIM Windows.....	22
5.4.2	POPs SIM Windows.....	22
6	Running a Set of Samples .....	24
6.1	Updating the Logbook.....	24
6.2	Preparing Standards.....	24
6.3	Setting Up a Sequence.....	25
6.4	Building a Sequence.....	27
6.4.1	Sequence Columns.....	27
6.4.2	Finishing and Saving the New Sequence .....	28
6.5	Running the Sequence .....	30
6.6	Calculating the Reproducibility Standards .....	30
6.7	Restarting a Sequence After an Error or the Run Has Been Aborted .....	31
6.8	Unloading the Autosampler Tray After the Run .....	32
7	MS Maintenance .....	33
7.1	Venting the MS .....	33
7.2	Pumping Down the MS .....	33
7.3	The Ion Source .....	34
7.3.1	Updating the Logbook.....	34
7.3.2	Removing the Source.....	34
7.3.3	Replacing the Source .....	35
7.3.4	Cleaning the Source.....	36
7.4	The Electron Multiplier (EM).....	40
7.4.1	Updating the Logbook.....	40
7.4.2	Removing the MS Analyzer .....	41
7.4.3	Installing a New Electron Multiplier.....	42
7.4.4	Installing the MS Analyzer .....	42

7.5	Adjusting the Radio Frequency.....	42
7.6	Changing the Rough Pump Oil.....	43
7.7	Refilling the Calibration Gas.....	43
8	Troubleshooting.....	44
8.1	Leaks.....	44
8.2	Signs That it is Time to Replace the Analytical Column .....	44
8.2.1	Checking Column Bleed.....	45
8.2.2	Peak Shape.....	45
8.2.3	Sensitivity.....	45
8.2.4	Noisy Baseline .....	45
8.3	Signs of a Dirty Ion Source.....	45
8.4	Checking/Replacing the Syringe .....	46
9	Helium .....	47
9.1	Helium Tanks .....	47
9.2	Helium Filters .....	47
10	Purchasing / Inventory .....	48
11	Sample Storage/Archival .....	49

## Figures

Figure 1. Agilent 7890A gas chromatograph, 7693 autosampler, and 5975C mass spectrograph .....	1
Figure 2. Oven control panel.....	2
Figure 3. Autosampler injector turret with rinse vials and syringe .....	2
Figure 4. Inlet septum cap.....	3
Figure 5. 1) Crescent wrench. 2) ¼" wrench. 3) Modified paper clip. 4) Forceps. 5) Magnifying loupe. 6) Sapphire cleaving tool. 7) Needle-nose pliers.....	4
Figure 6. 1) Inlet inserts. 2) Predrilled graphite ferrule. 3) Septums. 4) Column union. 5) Swaging nut. 6) Column nut. 7) Agilent "UltiMetal Plus" ferrule, 0.53 mm.....	4
Figure 7. GC oven, interior, with guard and analytical columns installed .....	4
Figure 8. Agilent "UltiMetal Plus" ferrules, 0.53 mm, for guard column.....	5
Figure 9. Column inlet nut.....	5
Figure 10. Agilent "UltiMetal Plus" ferrules, 0.25 mm, for analytical column.....	5
Figure 11. (top) An incorrectly cleaved column. (bottom) A correctly cleaved column .....	5
Figure 12. The column union connects the head of the analytical column to the end of the guard column.....	6
Figure 13. Leak detector.....	6
Figure 14. New Agilent guard column, 10 m × 0.53 mm.....	9
Figure 15. Torx T20 screwdriver tightening/loosening the column clip's screws inside the GC.....	10
Figure 16. Torx T20 screwdriver used to loosen/tighten the clip's screws that keep the column secured in the GC.....	11
Figure 17. New Agilent analytical column, 60 m × 0.25 mm .....	11
Figure 18. Replacement interface nut.....	11
Figure 19. Pre-drilled graphite ferrules for the interface .....	12
Figure 20. Measuring tool for the end of the analytical column that feeds into the MS .....	12
Figure 21. Three-hole die that cleans the interface threads before installation of the interface nut .....	12
Figure 22. AHP standard, properly prepared, labeled, level-marked.....	24
Figure 23. Vent valve knob. Also shown is the calibration gas bulb, filled with PFTBA (perfluorotributylamine) .....	33
Figure 24. View of ion source through the MS window .....	34
Figure 25. Top view (left) and side view (right) of the ion source inside the MS .....	35
Figure 26. Clean ion source secured in its protective case .....	36
Figure 27. Source cleaning tools.....	36

Figure 28. Disassembled ion source.....	37
Figure 29. Repeller.....	37
Figure 30. Ion source body.....	37
Figure 31. Draw-out cylinder (left) and plate (right) .....	38
Figure 32. Ion focus lens.....	38
Figure 33. Entrance lens .....	38
Figure 34. Transfer line socket.....	38
Figure 35. Side view of an assembled EI ion source.....	39
Figure 36. Top view of an assembled EI ion source.....	39
Figure 37. Side view of an assembled EI ion source.....	40
Figure 38. Side view of an assembled EI ion source.....	40
Figure 39. (left) Electron multiplier inside of the analyzer chamber. (right) Zoomed-in image of the electron multiplier.....	40
Figure 40. (left) Side image of the dark gray source power cable and the light gray control ribbon cable. (right) Front image of the source power cable and the sideboard ribbon control cable.....	41
Figure 41. Image of the panel door, open.....	41
Figure 42. Image of the HED control cable, the signal cable, and the HED power cable .....	42
Figure 43. Use these black screws to adjust the radio frequency.....	42
Figure 44. The oil level window on a GC/MS rough pump.....	43
Figure 45. Inside of the auto-sampler injector tower.....	46
Figure 46. Replacement syringe .....	46
Figure 47. (top) Helium gas tank bay for the GC/MS instruments. (bottom) Two open helium tanks hooked up to a switching valve.....	47
Figure 48. Helium gas filters for all four lines supplying gas to the GC/MSes .....	47

## Printout Examples

Example 1. An air/water spectrum printout, run to check the system for potential leaks.....	14
Example 2. A printout of an <b>stune</b> spectrum, run to assess the performance of the instrument .....	15
Example 3. A printout of an <b>atune</b> spectrum, run to assess the performance of the instrument.....	15
Example 4. A printout of an <b>sAHtune</b> spectrum, run to assess the performance of the instrument and ensure the sensitivity is appropriate .....	16
Example 5. A printout of a <b>CHtune</b> spectrum, run to assess the performance of the instrument and ensure the sensitivity is appropriate .....	17
Example 6. Suggested analytes to monitor in <b>AHP4A</b> standard, with acceptable peak shapes.....	20
Example 7. Suggested analytes to monitor in <b>CHBr2</b> standard, with acceptable peak shapes.....	21
Example 8. Suggested analytes to monitor for signs that it is time to update SIM window times in the POPs methods .....	23
Example 9. A printout of a sequence created to run a set of samples for PAHs analysis .....	29
Example 10. A printout of a sequence created to run a set of samples for POPs analysis.....	29

## Tables

Table 1. Standards used to run the POPs and PAHs analyses.....	25
Table 2. Standards, acquisition methods, and data processing methods and their corresponding matrix and target analyte lists .....	26

## Abbreviations and Terminology

AH	aromatic hydrocarbon	MS	mass spectrometry
AHP	AH standards with phytane	MSD	mass selective detector
AMU	atomic mass unit	NRDA	Natural Resource Damage Assessment
CB	chlorinated biphenyls	PAHs	polycyclic aromatic hydrocarbons
CHBr	chlorinated hydrocarbons brominated standards	PBDEs	polybrominated diphenyl ethers
DI water	deionized water	PCBs	polychlorinated biphenyls
EI	electron ionization	PFTBA	perfluorotributylamine (valve)
EM	electron multiplier	POPs	persistent organic pollutants
GC	gas chromatography	PPE	personal protective equipment
GC/MS	gas chromatography/mass spectrometry	PSEMP	Puget Sound Ecosystem Monitoring Program
HED	high-energy dynode	QA	quality assurance
HPCB	high molecular weight polychlorinated biphenyls	QC	quality control
HPLC	high performance liquid chromatography	Repro.	reproducibility (standard)
LCREP	Lower Columbia River Estuary Partnership	RSD	relative standard deviation
LVI	limited volume insert	SIM	selective ion monitoring (mode)
		SRM	standard reference material

### GC/MS systems

- #1:** Set up to run POPs analysis.
- #5:** Set up to run PAHs analysis.
- #6:** Set up for method development/POPs analysis.
- #7:** Set up to run POPs analysis.

### Commonly used GC buttons

- AuxTemp:** Displays the current temperature of the auxiliary.
- Col 1:** Displays the column that is installed in the Column 1 position.
- P:** Parks the autosampler.
- Oven:** Displays the temperature and column flow for the GC oven.
- Time:** Displays the run time and the time remaining until the next injection.

### Tune files

- sAHtune:** Standard aromatic hydrocarbon tune (used for PAHs analysis).
- stune:** Standard tune (used for PAHs analysis).
- atune:** Auto-tune (used for POPs analysis).
- CHtune:** Chlorinated hydrocarbon tune (used for POPs analysis).

## Acknowledgments

The authors would like to extend a special thank you to Jennifer Travis (Northwest Fisheries Science Center) for her help with the photography throughout this Processed Report, and to Su Kim (NWFSC) for her assistance with the creation of the title page.

# 1 Introduction

Gas chromatograph/mass spectrometer (GC/MS; Figure 1) systems are instruments used to determine the concentrations of contaminants in a given sample. These systems are complex and sensitive, and require detailed care and regular maintenance to maintain a high level of performance. This comprehensive protocol is written to be used concurrently with hands-on training. Some steps require more feel and familiarity than can be described here and, as such, are acquired with practice.

This protocol provides step-by-step instructions for everything from maintenance to loading samples for analysis. Both the persistent organic pollutants (POPs) and the polycyclic aromatic hydrocarbons (PAHs) analyses will be discussed in detail throughout this document. Full-color photographs, computer images, and multiple examples will make the instructions clearer.

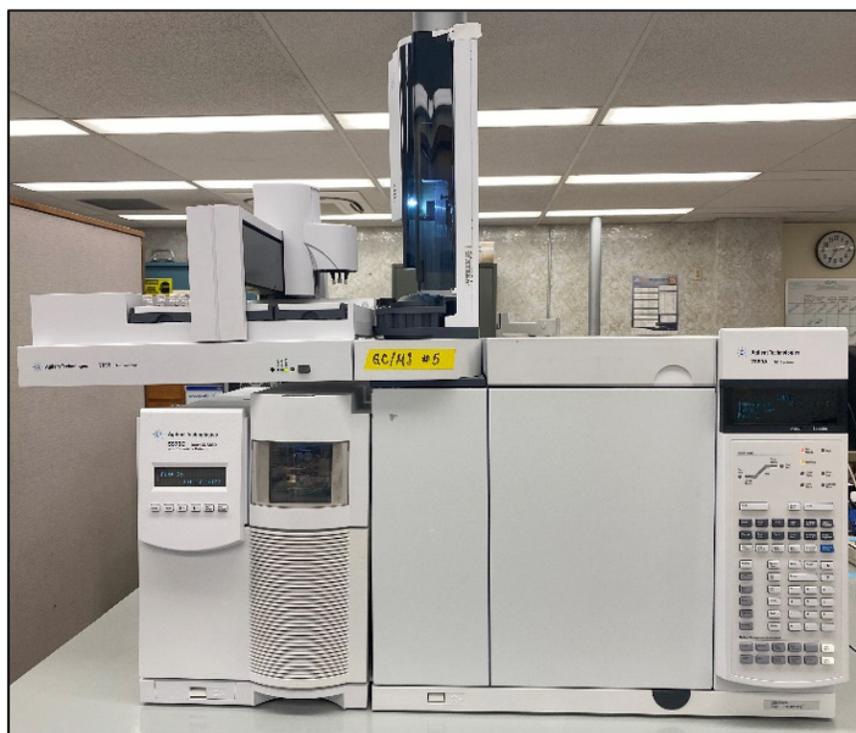


Figure 1. Agilent 7890A gas chromatograph, 7693 autosampler, and 5975C mass spectrophotometer.

## 2 Routine Maintenance

Routine maintenance should be performed when peak shape and/or sensitivity begin to decline. Routine maintenance consists of cleaning the inlet and trimming the guard column. Using the control panel on the GC (Figure 2), cool the oven to 30°C and turn the **Aux Temp** (interface heater) off. Remove the auto-sampler injector tower (Figure 3) and mount it on the post on top of the GC at the back, then lift the top of the GC to facilitate cooling and provide access to the top of the inlet. The GC will take approximately half an hour to cool down (the interface heater needs to be below 100°C), but temporarily increasing the column helium flow to 3 mL/min will decrease this time.

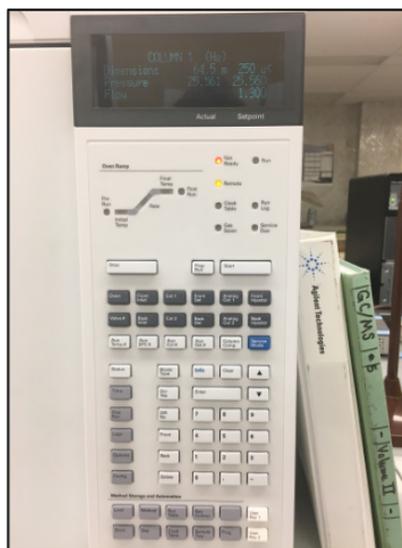


Figure 2. Oven control panel.

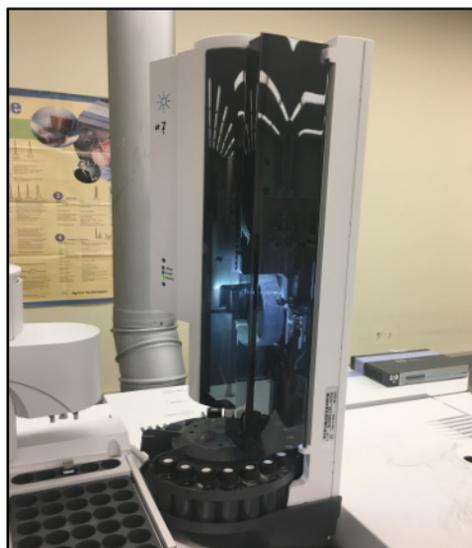


Figure 3. Autosampler injector turret with rinse vials and syringe.

### 2.1 Updating the Logbook

All maintenance needs to be recorded in the instrument's logbook. Highlight these entries so they can easily be found months down the line. Generally, looking at previous maintenance logs will give the writer a template for recording completed maintenance.

*Basic information required:*

- Whether the GC and/or MS was cooled.
- The amount trimmed off the guard and/or analytical column and the amount of column remaining.
- Whether the guard or analytical column was replaced (include the serial number of the new column if applicable).
- Replacing the inlet insert and septum.
- Whether fittings were checked for leaks.
- Helium flushing.
- Equilibrating.

Any anomalies must also be recorded (for instance, if a column spontaneously breaks inside the union). All entries should be signed or initialed. Throughout this standard operating procedure, important actions have been highlighted to remind the reader to update the logbook after maintenance has been performed.

*Examples of logbook entries for routine maintenance:*

- “Cooled GC. Trimmed 1 m from head of guard column (7 m left). Installed clean inlet insert and new septum. Allowed columns to helium-flush and checked fittings for leaks. Up-heated system and allowed to equilibrate.”
- “Cooled GC. Removed remainder of existing guard column and replaced with new Agilent 10-m × 0.53-mm guard column. Installed clean inlet insert and new septum. Allowed columns to helium-flush and checked fittings for leaks. Up-heated system and allowed to equilibrate.”

## 2.2 Cleaning the Inlet

Once the GC has cooled, the helium flow must be turned off. Press **Col 1** and turn the pressure off. Open the GC oven door and, using a ¼" open-end wrench, loosen the column inlet nut from the bottom of the inlet and remove the column from the fitting. Now cleave the column just behind the ferrule so the ferrule and column nut can be removed from the end of the column. Point the open end of the column down while trimming the column to avoid particles falling into the column. Disassemble the top of the inlet using needle-nose pliers and forceps. Use the pliers to unscrew the septum cap (Figure 4) and the forceps to remove the septum and the spring. Discard the old septum and temporarily place the spring inside the septum cap, so as not to lose it. Now remove the dirty inlet insert by inserting a straightened paper clip or the end of the guard column into the bottom of the inlet and pushing up and out. Before reassembling the inlet, it is a good idea to give it a shot of canned air into the top and the bottom to remove any fragments of column or septum that may have accumulated. (*Note:* Do not shake the can or hold it upside down.) Next, working at the top of the inlet, install a clean insert and replace the spring on top of the insert. Put a new septum in the cap (with the indent facing the top of the cap) and screw the septum cap snugly back on the inlet. Dirty inlet inserts are cleaned for reuse (see [Section 2.5](#)). See Figures 5 and 6 for tools and GC parts.



Figure 4. Inlet septum cap.

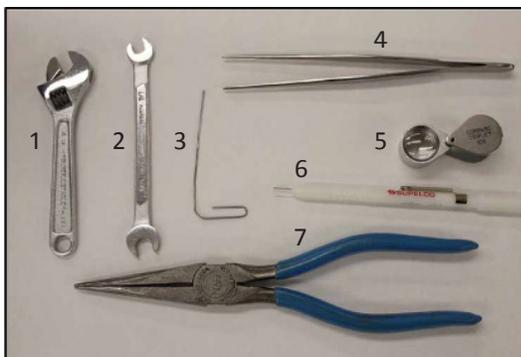


Figure 5. 1) Crescent wrench. 2) 1/4" wrench. 3) Modified paper clip. 4) Forceps. 5) Magnifying loupe. 6) Sapphire cleaving tool. 7) Needle-nose pliers.

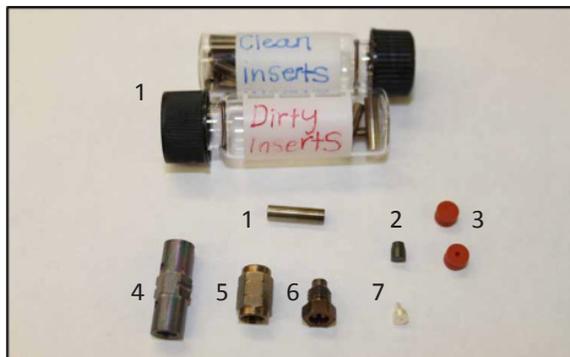


Figure 6. 1) Inlet inserts. 2) Predrilled graphite ferrule. 3) Septums. 4) Column union. 5) Swaging nut. 6) Column nut. 7) Agilent "UltiMetal Plus" ferrule, 0.53 mm.

## 2.3 Trimming the Guard Column

When peak shape and sensitivity have deteriorated, half a meter should be trimmed from the head of the guard column. After every 2–3 sets of PAHs or 4–5 sets of POPs, depending on the type and condition of the matrix, the analytical column will usually require trimming. Once the guard column has run sample sets at 4 meters in length, then it is replaced. Likewise, when the analytical column has run sets at 53 meters, it is replaced (Figure 7).

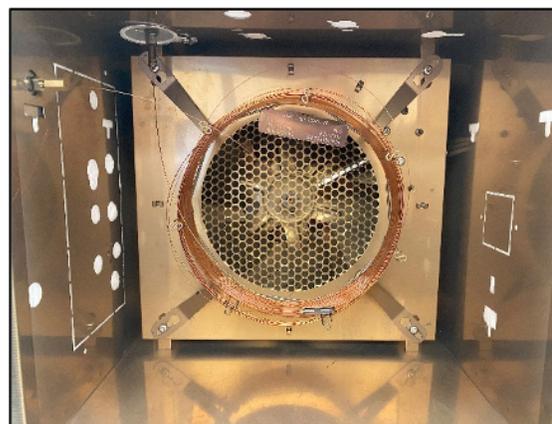


Figure 7. GC oven, interior, with guard and analytical columns installed.

### 2.3.1 Guard Column Maintenance

When inlet maintenance is complete, guard-column maintenance is performed. This requires a sapphire column cleaving tool, meter stick, magnifying loupe, 1/4" wrench, and a 0.53-mm Agilent "UltiMetal Plus" ferrule (Figures 5, 6, and 8). You should already have removed the guard column from the inlet during inlet maintenance. Unwind the column as necessary and use the cleaving tool and the meter stick to trim approximately 0.5 meters from the head of the guard column. Point the open end of the column down to avoid particles falling into the column. Use the magnifying loupe (Figure 5) to ensure the cut is smooth and perpendicular (Figure 11). Discard the trimmed portion of the column. Now slide the column inlet nut (Figure 9) back onto the end of the column (threads facing the end), followed by the abovementioned ferrule (tapered end facing the end of the column). Insert the end of the column up into the inlet until you feel the column stop, then finger-tighten the nut into the inlet without letting go of the column. While still holding the column in place, use the 1/4" wrench to further tighten the nut into the fitting (usually no more than ~1/4 turn). Take care not to over-tighten the nut, as this can cause the column to shear off (the guard columns are very susceptible to this). Check the fittings for leaks (Section 2.3.3), and update the column length in the methods (Section 2.3.4).



Figure 8. Agilent "UltiMetal Plus" ferrules, 0.53 mm, for guard column.



Figure 9. Column inlet nut.



Figure 10. Agilent "UltiMetal Plus" ferrules, 0.25 mm, for analytical column.

### 2.3.2 Analytical Column Maintenance

Analytical column maintenance is generally performed after inlet and guard column maintenance have been completed. This is not part of routine maintenance, but should be performed after every 4–5 sets for systems running POPs or every 2–3 sets for systems running PAHs, as needed depending on the type and condition of the matrices. Analytical column maintenance should also be performed if there is a problem with peak shape and trimming the guard column doesn't resolve the issue. You will need a 0.25-mm Agilent "UltiMetal Plus" ferrule (Figure 10), the  $\frac{1}{4}$ " open-end and crescent wrenches, a swaging nut, and a meter stick (Figures 5 and 6). Using the wrenches, loosen and remove the analytical column from the union. Unwind enough of the column to trim off approximately 1 meter. Point the open end of the column down while trimming to avoid particles falling into the column. Use the magnifying loupe to ensure the cut is smooth and perpendicular (Figure 11). Also make sure that the end of the column will meet up with the union. Discard the trimmed portion of the column. Place the union nut (Figure 12), threads facing the end of the column, back onto the column, followed by a new 0.25-mm ferrule (tapered end facing the end of the column). Next, screw the swaging nut (Figure 6) onto the column union nut. Once again, tighten the swaging nut just enough that the ferrule stays in place but can still be moved on the column without stripping off the outer coating of the column. Unscrew and remove the

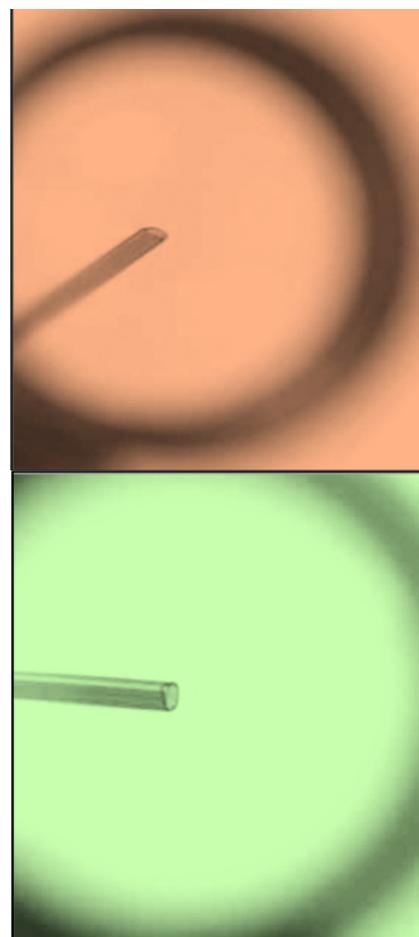


Figure 11. (top) An incorrectly cleaved column. (bottom) A correctly cleaved column.

swaging nut, then position the ferrule so that approximately 2 cm of the column protrudes. Now gently fit the column back into the union (this may take several attempts, as the analytical column must actually be inserted inside the guard column). Tighten the union nut pretty snugly, but take care not to over-tighten it (although the analytical columns are much less likely to break than the guard columns). Check the fittings for leaks ([Section 2.3.3](#)), and update the column length in the methods ([Section 2.3.4](#)).

You may need to bake out the system if you have an old analytical column. If there are extra peaks in your chromatograms, try turning the oven temperature up to 320°C and the flow up a little bit (making sure not to let the pressure exceed 50 psi). Let the system sit there for 3–4 hours.

### 2.3.3 Checking Fittings for Leaks

After opening and refitting any column fittings in the GC, the fittings must be checked for helium leaks using the electronic leak detector. Once all fittings are properly tightened, press **Col 1** on the GC control panel and set the flow to 3 mL/min. Remove the leak detector (Figure 13) probe from the unit, then turn the leak detector on using the power button and allow it to settle. If there are any residual red dots, press **Reset** and allow it to stabilize. Place the probe next to each fitting for about 30 seconds while monitoring the detector for signs of a leak. For the inlet fittings, check the top and bottom of the inlet nut. For the union, check both sides of the union. It is important to leak-check the spot at which the nut screws into the union as well as the back end of the nut where the column comes out. If there is a leak, the detector will begin to light up with red indicator lights and make an audible alarm. Sometimes there is a small amount of background gas that will light up one or two indicator lights; press **Reset** to clear it out and recheck the fitting to see if it still persists. If there is a leak, the fittings should be tightened slightly until fixed. If tightening the fittings isn't fixing the leak: 1) cool the **Aux Temp** below 100°C (if not cooled already), 2) open the fittings (the flow/pressure should be turned off if the inlet is opened up), 3) inspect them for problems (e.g., improperly assembled inlet or union, bad septum, broken column), 4) reassemble, and 5) recheck for leaks.



Figure 12. The column union connects the head of the analytical column to the end of the guard column.



Figure 13. Leak detector.

### 2.3.4 Updating Column Length in Methods

Column length must be updated in Acquisition and Conditioning Methods after the guard column and/or the analytical column is trimmed or replaced and before tuning and running standards. The methods that need to be updated for PAHs instruments are: **condn.m**, **AHscan.m**, and the PAHs method to be used for the next analyses (depending on matrix and analytes): **AHSIM.m**, **AlkSIM.m**, **AlkSIM2.m**, or **AlkSIM3.m**. The methods that need to be updated for POPs instruments are: **CHBrSim.m**, **CHBrStd.m**, **condn.m**, and **CHBrwndw.m**. In the ChemStation **Instrument Control** window, load the appropriate method. Click on the column icon button, then click on the column icon button in the **Parameters** pop-up window if it isn't already selected. If the instrument you are working on has newer software and there isn't an inventory, click **Change Column**, then edit the length (total length), model number, and description in the **Edit Properties of Installed GC Column** pop-up window. For GC/MS #1, click on the column icon button then click on the column icon button in the next pop-up window (**Instrument | Edit | Oven:(6890)**) if it isn't already selected. Click **Change**, then **Install**. Select the appropriate column from the inventory list if it is present, then click **OK** and the next two **OKs**, and save the method. If the appropriate column isn't present in the inventory list, click **Add**, then enter the new column's description in the box for **Inventory # for New Column**, and click **OK**. In the next window, click **Add New Column Model To Catalog** and fill in the fields appropriately.

Below is an example of a new column length that needs to be added to the catalog (in this example, for a 54-m DB-5 analytical column plus an 8.5-m × 0.53-mm guard column) and the responses you must enter into the system in order for the software to recognize the overall change in column length:

**Manufacturer:** J&W

**Model #:** DB54+8.5

**Description:** 54-m DB-5 + 8.5-m × 0.53-mm guard

**Column Type:** capillary

**Max Temperature:** 350°C

**Capillary Column Dimensions:** Length: 62.5 m; Diameter: 250 mm; Film Thickness: 0.25 mm

After editing the column information, click **OK** and the next **OK**, then **Install as Column 1** and the next two **OKs**. Save the method.

Older software will alert you to the change the column length when you load another method, then update it for you in the method (remember to save the method). The newer software will not alert you. If the software does not give an alert when you load another method, repeat the process for editing the column information and save each method until all methods to be used are updated.

## 2.4 Changing the Rinse Vials

The rinse vials in the autosampler injector turret (Figure 3) need to be changed periodically. Check them before, during, and after each sample sequence to make sure the level of the solvent is never below the 3-mL mark on the 4-mL rinse vial. The first six rinse vials contain 100% methanol and the last four vials contain a 50:50 solution of dichloromethane:isooctane. If a 50:50 rinse vial is at or around the 3.5-mL mark, it may be topped off with isooctane from the 1-L pumper bottle. Once a 50:50 rinse vial has been topped off, use a marker to mark the bottom of the vial. When a marked rinse vial has become low, it is time to replace it with a new one. Never top off a rinse vial more than once. When the level of the 100% methanol rinse vials is below the 3-mL mark, it is time to replace it with a full 100% methanol rinse vial. New rinse vials are located next to the sink in **Room 317E**.

## 2.5 Cleaning the Inlet Inserts

The inlet insert (Figure 6) is removed and replaced with a clean insert whenever inlet maintenance is performed ([Section 2.2](#)). Dirty inlet inserts are sonicated in methanol, then acetone, then methylene chloride for 10 minutes each, draining and disposing of each solvent in between. Proper use of personal protective equipment (PPE) and a fume hood is required while working with solvents. When the inserts have gone through all three steps and the methylene chloride has been drained, remove them with clean forceps and place them in the **Clean Inserts** 4-mL vial. Put the vial in the fume hood, without a lid, to fully dry (approximately 30 min).

## 3 Replacing the Columns

Once the guard column has run a sample set at 4 meters in length, then it is replaced. Likewise, when the analytical column has run a set at 53 meters, it is replaced.

### 3.1 Updating the Logbook

When replacing columns, it is important to record everything in the logbook. It is useful to keep track of how long a guard or analytical column has been installed, especially when troubleshooting issues with the system down the road. *Key details include:* the make and model of the new column(s), the new column's length and lot number, and a notation that the new column's length was updated within the methods. It is also important to keep the original boxes of the columns near the MS in case there is an issue after installation. Highlight this entry so it can easily be found months down the line.

*Examples of logbook entries for replacing columns:*

- “Cooled GC. Removed the remainder of the guard column and replaced it with a new UltiMetal Plus 10-m × 0.53-mm guard column (#5030880). Installed a new inlet septum and clean insert. Allowed columns to He-flush while checking fittings for leaks, then up-heated the system and allowed to equilibrate.”
- “Removed existing analytical column and installed a new Agilent/J&W 60-m × 0.25-mm DB-5 analytical column (#USN685845H) to the new guard column (60 + 10). Allowed columns to He-flush while checking fittings for leaks, then up-heated the system and allowed to equilibrate then began conditioning the columns.”

### 3.2 Replacing the Guard Column

As mentioned above, once the guard column has run a sample sequence at 4 meters in length (or if there is a problem with the column), it should be replaced. Cool the GC (oven to 30°C, **AuxTemp** turned to **OFF**). Once the GC is cool, turn the helium flow off (**Col 1** pressure **OFF**). Unscrew the column inlet nut (Figure 9), lower the existing guard column from the GC inlet, then cut off the end of the column to remove the nut and ferrule. Next, unscrew the guard column from the union (Figure 12) and cut off that end of the column to remove the nut and ferrule (Figure 6). Also, unscrew the analytical column from the union and inspect the end. If the end has broken off at the tip, it must be trimmed and refitted with a new ferrule. Prepping the new guard column (Figure 14) requires a new 0.53-mm Agilent “UltiMetal Plus” ferrule (Figure 8), the ¼” wrench, the swaging nut, the union nut, and the union body (Figures 5 and 6). The structure of the union body is the same at both

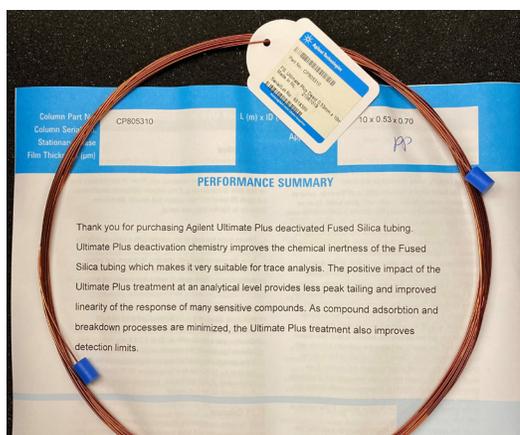


Figure 14. New Agilent guard column, 10 m × 0.53 mm.

ends, so either end of the union will accommodate either column. Trim approximately 5 cm off each end of the new guard column while pointing the open end of the column down to avoid particles falling into the column. Use the magnifying loupe to ensure the cut is smooth and perpendicular (Figure 11). Place the union nut (threaded end facing out) and a new 0.53-mm ferrule on one end of the column. Tighten the swaging nut onto the union nut using the wrench; do not over-tighten. (This is a bit tricky and usually requires some practice, as the ferrule must remain moveable on the column without stripping off the outer coating of the column, but stay in place once positioned). Remove the swaging nut. Position the ferrule at the end of the column so that only approximately 1.5 mm extends from the tip of the ferrule. Now fit the guard column into the union. Position the column in the GC so the union will line up with the end of the analytical column, and reinstall the analytical column in the union (see [Section 2.3.2](#)). Insert the other end of the guard column into the inlet ([Section 2.3.1](#)). Check the fittings for leaks ([Section 2.3.3](#)). Once no leaks are detected, allow the system to helium-flush for an hour; then reduce the helium flow to 1.3 mL/min and up-heat the GC by turning the oven to 120°C and the **AuxTemp** back on (to 320°C). Allow the system to equilibrate (the time required here is highly variable; sometimes it takes only a couple hours, other times overnight is needed). Monitoring the air/water spectrum and the system background is key here. (For help, proceed to [Section 4.2](#).) Update the column length in the methods ([Section 2.3.4](#)).

### 3.3 Replacing the Analytical Column

The analytical column needs to be replaced when the length gets down to 53 meters, if the column bleed is much higher than when you initially installed it ([Section 8.2](#)), or if there are any other suspected issues that are not being resolved with routine maintenance. Begin with putting the system in vent mode by clicking **View** and selecting **Tune and Vacuum Control**, then clicking **Vacuum** and selecting **Vent** in ChemStation **Instrument Control**. This will turn off the turbo pump and start cooling the MS, generally taking about an hour to complete. It will also automatically shut the auxiliary heater and the oven off, so manually set the oven temperature back up to 30°C. Once the MS is cool and the turbo pump has stopped, the **Instrument Control** software will shut itself down.

Remove the existing analytical column, starting with the interface. Next, cap the interface off with a plugged interface nut. Remove the whole spool of columns by loosening the clip's screws (Figure 15) using the red-handled Torx T20 screwdriver (Figure 16). If the guard column is down to 4–5 meters, you can toss it as well and replace it with a new one. When replacing an analytical column (Figure 17), the interface nut will need to be replaced (Figure 18) as well, because tightening over time causes them to get distorted. Before tossing everything, make sure all the hardware (like the nuts and union) is removed. Remove approximately 10 cm off of the head of the analytical column (the head is the end toward the back of the spool) while pointing the open end of the column down to avoid particles falling into the column. Use the magnifying loupe to ensure the cut is smooth and perpendicular (Figure 11). The head will be



Figure 15. Torx T20 screwdriver tightening/loosening the column clip's screws inside the GC.



Figure 16. Torx T20 screwdriver used to loosen/tighten the clip's screws that keep the column secured in the GC.

joined to the guard column. Start by installing the nut onto the end of the guard column using a swaging nut, then install the union and tighten. Once the union is installed on the guard column, install the nut on the head of the analytical column using a swaging nut. Join the two columns together with the union and tighten. It is now time to put the spool of columns back into the GC. Place the spool in so that the head of the guard column can fit all the way into the inlet. Next, attach the clips and tighten up the screws with the red-handled Torx T20 screwdriver. Place the column in the inlet and tighten. Make sure to analyze the inlet nut to make sure the threads are in good shape. Remove the blue septum from the end of the analytical column and set the helium flow to 3 mL/min in the GC. Remove approximately 5 cm off the end of the column to ensure that it isn't plugged with the septum. Using a rinse vial, put the open end of the analytical column into the rinse vial to see if there are lots of bubbles flowing. Use the leak detector to check the fittings for leaks ([Section 2.3.3](#)), making sure to keep the detector away from the open end of the spool. Update the column length in the methods ([Section 2.3.4](#)). Let the column flush with the helium at a flow of 3 mL/min for approximately an hour, then condition the analytical column ([Section 3.3.1](#)).

### 3.3.1 Conditioning a New Analytical Column

To condition a new analytical column, increase the oven temperature from 30°C to 320°C in 40-50°C increments. (The first increase should be from 30–80°C and the second from 80–120°C. From there, increase by 50°C until 320°C is reached.) Hold for approximately 30 minutes between each temperature change until 320°C is reached, then let it sit for 3–4 hours. You can leave the flow at 3mL/min at 80°C, but when the temperature is increased to 120°C the flow will have to be decreased to keep the pressure under 50 psi! There is not enough time in one day to reach all of the temperatures for conditioning, so the oven should be set at 120°C with a flow of 1.3 mL/min overnight. In the morning, resume the column conditioning at the next increment in the conditioning process, with an appropriate flow to keep the pressure under 50 psi.

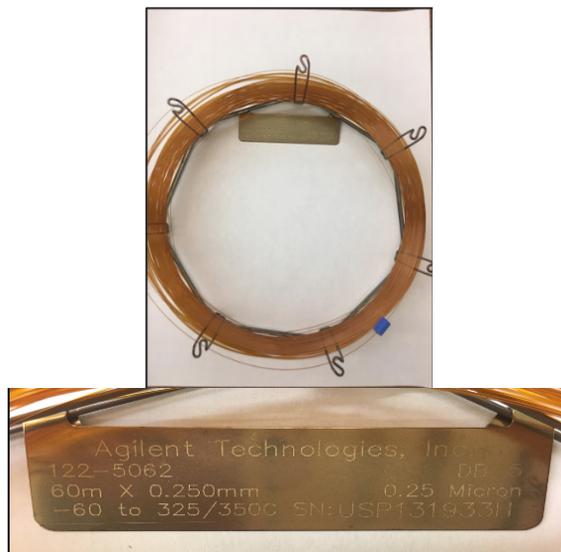


Figure 17. New Agilent analytical column, 60 m × 0.25 mm.



Figure 18. Replacement interface nut.

### 3.3.2 Installing the Analytical Column in the Mass Spectrometer

Once the column has been conditioned, cool the oven down to 30°C (from 320°C) and set the flow to 1.3 mL/min. Place a new interface nut (Figure 18) on the end of the column and place your predrilled graphite ferrule (Figure 19) on backwards (small end first). Trim approximately 10 cm off of the end of the analytical column while pointing the open end of the column down to avoid particles falling into the column. Using canned air, blow off the end of the analytical column. (Do not shake the can or hold it upside-down.). Put the analytical column into the analytical column tool (Figure 20) until it sticks out the end of the tool, and start tightening the nut onto the tool using two wrenches until it starts getting harder to pull the column through the tool. Next, using a ruler, back it off until you have 1–1.5 mm sticking out of the tool. When the column is positioned properly, tighten the nut to the tool as tightly as possible. Now mark where the analytical column meets the nut with Wite-Out in case the column slips when the tool is removed. Take the plug off of the interface. Using the three-hole die (Figure 21), clean the threads off of the interface and give it a shot of canned air. Now, using the wrenches, take the column out of the measuring tool and put it into the interface. Tighten up the nut until it's snug without over-tightening, making sure the column did not move. At this point, make sure all of the inlet and union fittings are tight. (When the oven spends a lot of time at 320°C and then is cooled, things tend to loosen up). Now increase the flow to 3 mL/min and check for leaks; if there are no leaks, turn the flow down to 1.3 mL/min and turn the oven to 80°C. Restart ChemStation **Instrument Control** on the computer (this will take a few minutes). Click **OK** on the temperature window that pops up. Once the program is started, go to **Abort** and click **OK**. Go to **View**, then **Tune and Vacuum Control**. Click **OK** again when the temperature window pops up. Go to **Vacuum**, then **Pump Down**. Click **OK** on all the windows that pop up. The **MS Source Target** temperature should be 300°C and the **MS Quad Target** temperature should be 150°C. **Instrument Control** should reheat the ovens, but check the GC control panel to make sure. The system needs to pump down for a minimum of 4 hours before the air/water spectrum is checked ([Section 4.2](#)), but for best results it is ideal to allow the system to pump down overnight before running any tests.



Figure 19. Pre-drilled graphite ferrules for the interface.



Figure 20. Measuring tool for the end of the analytical column that feeds into the MS.



Figure 21. Three-hole die that cleans the interface threads before installation of the interface nut.

### 3.3.3 Checking the New Column's Column Bleed

After installing the new analytical column, run an AHP7 standard on the instrument using the **AHscan** method. When the standard is done running, zoom the baseline in on the end of the run and print out the chromatogram. The chromatogram should be placed inside of the new analytical column box beside the instrument so that it can be used to compare the bleed months after installation. Anything under 80,000 counts is acceptable. When you first install the column it should be low, around 15,000 or 20,000 counts.

## 4 Tuning the Mass Spectrometer

Tune the MS after performing maintenance, after the system has sat idle for a long period of time, and in between sample set runs, if needed. Once the system has equilibrated after maintenance and the column length in the acquisition method is current, the air/water spectrum needs to be checked to confirm there are no leaks. Then, tune the system using auto-tune (**atune**) for POPs or standard spectrum tune (**stune**) for PAHs. Adjust the tune and save it as **CHtune.u** (for POPs) or **sAHtune.u** (for PAHs), and scan the air/water spectrum using the new **atune** or **stune** parameters. While tuning and checking the air/water spectrum, the oven temperature should be 120°C, the auxiliary temperature should be on (320°C), and helium flow should be 1.3 mL/min.

### 4.1 Updating the Logbook

Tuning requires a logbook entry. Look at previous tune entries for an idea about how they are usually written. Basic information to include in a tune entry: status of the air/water spectrum, background levels, and stability of mass abundances.

*Example of a logbook entry for tuning:*

- “Checked out/retuned system. Everything looks OK. I bumped up the EM in both tunes significantly to gain sensitivity.”

### 4.2 Air/Water Spectrum

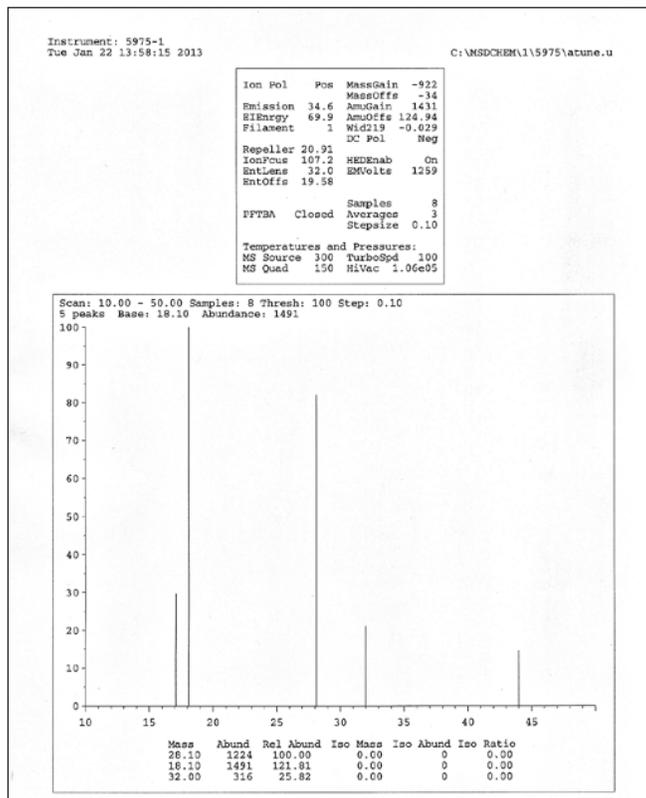
It is vital to check the air/water spectrum before and after tuning the MS. The transfer line oven temperature should be at 320°C, and the oven temperature set to 120°C at least five minutes prior to checking the air/water spectrum. Check the air/water spectrum by monitoring masses 28 (nitrogen), 18 (water), and 32 (oxygen) in **atune** (for systems analyzing POPs) or in **stune** (for systems analyzing PAHs) with the perfluorotributylamine (PFTBA) valve closed and the **Scan Range** set from 0 to 50.

#### 4.2.1 Checking the Air/Water Spectrum Before Tuning

In the MS control window, click **View**, then select **Tune/Vacuum Control**. Under **File**, load the file **atune.u** or **stune.u** depending on what you are running on that instrument (**atune** for POPs, **stune** for PAHs). Click **Parameters**, then select **Manual Tune**, which will show all the parameters of the tune. If the **Tune & Display Parameters** window doesn't open next to the **Manual Tune Parameters**, click **MoreParams** in the **Manual Tune Parameters** window and select **Tune Params**. To check the air/water spectrum, close the PFTBA valve, change the three masses to 28, 18, and 32 respectively, change the **Scan Range** to 0–50, and then click **Scan**. A spectrum will show the abundances of the three masses (nitrogen, water, and oxygen). If the abundance of nitrogen is over 3,000, the system may need to sit longer to equilibrate. If the abundance of nitrogen and oxygen is well over 3,000 (>7,000), there might be a leak in the system. Cool the oven to 30°C, then open up the GC and tighten the inlet and MS interface fittings. Heat the oven back up to 120°C and check the spectrum again. When the nitrogen is <3,000 and water and oxygen are <2,000, the system can be tuned. When exiting **Tune/Vacuum Control**, if a pop-up window asks if you want to use the tune file for the method, click **No**.

## 4.2.2 Checking the Air/Water Spectrum After Tuning

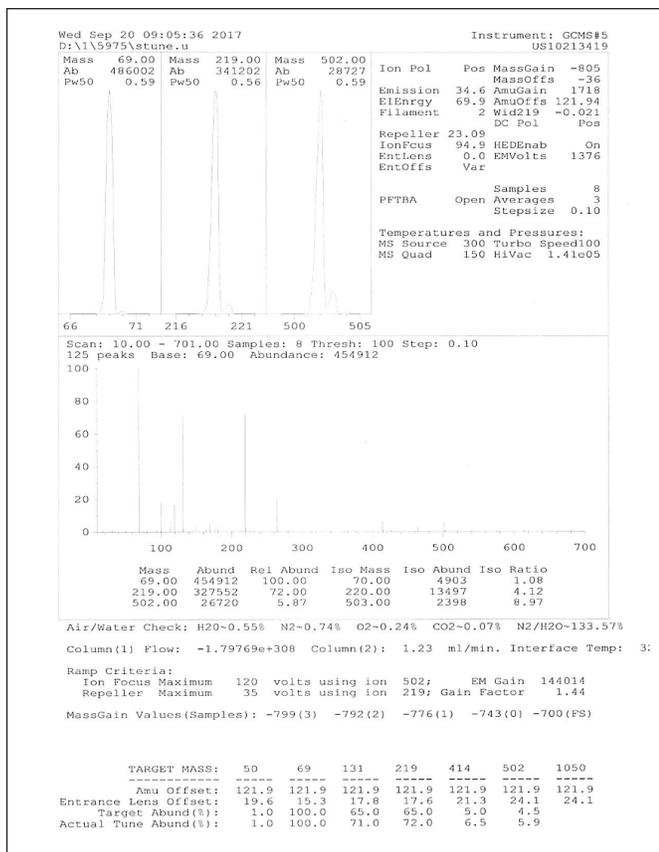
After tuning the system (Section 4.3), be certain the **CHtune** or **AHstune** file is saved and reload the **atune** or **stune** file to check the air/water spectrum. Click **Parameters**, then select **Manual Tune**. Close the PFTBA valve, change the three masses to 28, 18, and 32, respectively, change the **Scan Range** to 0–50, and then click **Scan**. Try for an average abundance for nitrogen, then click **Stop**. Click **File**, then select **Print** to print the spectrum for the tune binder. (Example 1) When exiting **Tune/Vacuum Control**, if a pop-up window asks if you want to use the tune file for the method, click **No**.



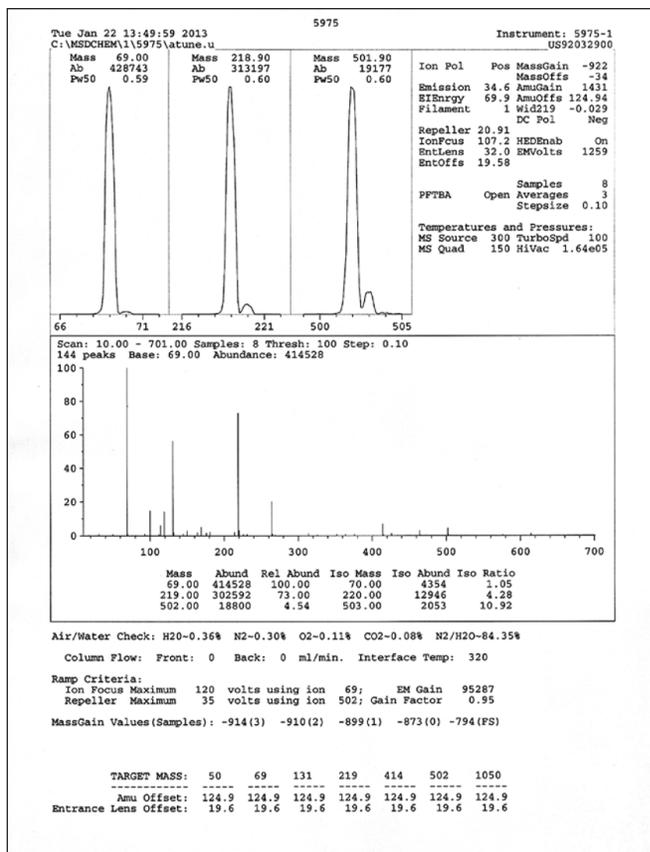
Example 1. An air/water spectrum printout, run to check the system for potential leaks.

## 4.3 Auto Tune (atune) and Standard Spectrum Tune (stune)

**atune** is the initial tune for systems that analyze samples for POPs; **stune** is the initial tune for systems that analyze samples for PAHs. In the **Tune/Vacuum Control** screen, load the file **atune.u** or **stune.u** and open **Manual Tune Parameters**. The windows that appear should not need any adjustment, except to open the PFTBA valve (if closed) and set the **Scan Range** to 0–700. The **EM Volts** should be those saved at the last tune's value. Click **Scan**. The spectrum will show the abundances of the masses. The lowest mass (69) should be about 420,000 counts. The background should be around 100–150 peaks. Click **Tune** and select **Tune MSD**. The computer will go through a variety of screens as the MS tunes; this will take a few minutes. The final window will show abundances for three masses, and the tune report will be printed. The new **atune.u** or **stune.u** file is saved automatically. Click **File**, then select **Generate Report** (Examples 2 and 3). This generates and prints another tune report using the same parameters, the purpose being to check instrument stability. If the 69 abundance fluctuates wildly, the system may need to sit and equilibrate for a few hours. If the abundances remain stable, then choose the printout that best represents where the system is (the average abundance) and put it into the tune binder.



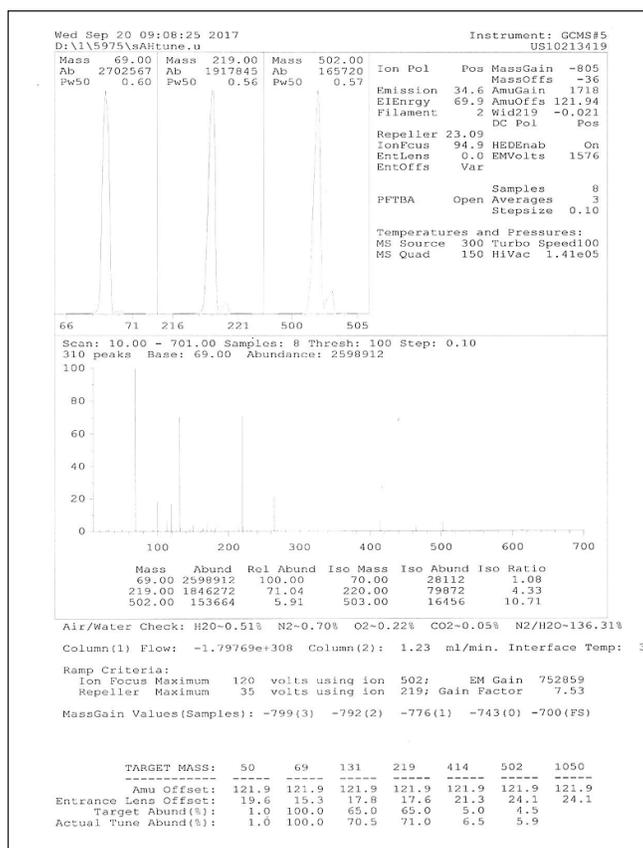
Example 2. A printout of an **stune** spectrum, run to assess the performance of the instrument.



Example 3. A printout of an **atune** spectrum, run to assess the performance of the instrument.

## 4.4 Standard Spectrum Tune File Used for PAHs Analysis (sAHTune)

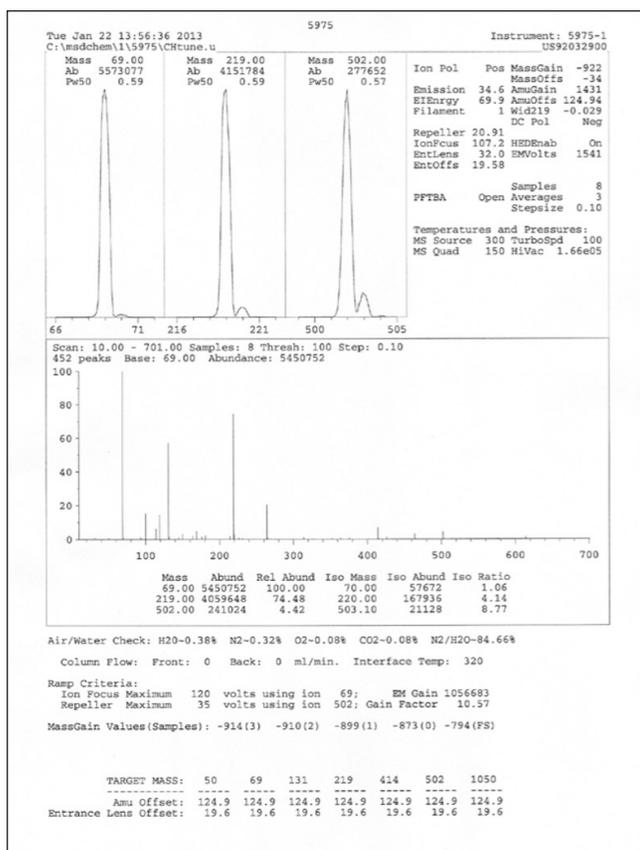
Go to the manual tune screen while still in **stune**. Turn the voltage on the EM to the value from the last **sAHTune.u** file, found in the tune binder, then adjust the EM voltage to achieve the desired abundance: the 69 mass in **sAHTune** should be around 2,000,000 counts. The 502 should be at least 5% relative abundance. Save the tune as **sAHTune**. Generate at least three reports, or until the abundances have stabilized (Example 4). If the abundances continually fluctuate, use the spectrum scan to save paper while checking stability. If the abundances remain stable, choose the printout that best represents where the system is at (average abundance) and put it into the tune binder. If the EM voltage must be changed at any point, remember to save the method.



Example 4. A printout of an **sAHTune** spectrum, run to assess the performance of the instrument and ensure the sensitivity is appropriate. The methods used to run PAHs analysis use **sAHTune**.

## 4.5 Tune File Used for POPs Analysis (CHtune)

Go to the manual tune screen while still in **atune**. Adjust the voltage on the EM to achieve the desired abundance, using the previous **CHtune** EM voltage as a guide to start: the 69 mass in **CHtune** should be around 5,000,000. The 502 should be at least 3% relative abundance. Save the tune as **CHtune**. Generate at least three reports, or until the abundances have stabilized (Example 5). If the abundances continually fluctuate, use the spectrum scan to save paper while checking stability. If the abundances remain stable, choose the printout that best represents where the system is at (average abundance) and put it into the tune binder. If the EM voltage must be changed at any point, remember to save the **CHtune** file before exiting the **Tune/Vacuum Control** screen.



Example 5. A printout of a **CHtune** spectrum, run to assess the performance of the instrument and ensure the sensitivity is appropriate. The methods used to run POPs analysis use **CHtune**.

## 5 Running Standards Post-Maintenance

Standards are run to assess performance of the instrument as well as to update SIM window times. Check the chromatograms of the standards for peak shape and sensitivity.

*Example of a logbook entry for instrument performance checks/updating SIM windows:*

- “Standards run over the weekend show good peak shape and HIGH sensitivity. I updated SIM window times in CHBr methods. System is ready to run samples.”

### 5.1 Choosing Standards

PAHs instruments always run **AHP1A** and **AHP4A** using the SIM method, which will be used for the next sample sets to be analyzed. If SIM windows need to be updated, such as after major maintenance has been performed (e.g., replacing one of the columns), then **AHP7A** should also be run using **AHscan.m**. Depending on the samples that will be run next on the instrument, the PAHs standards should be run with a corresponding conditioning solution. Instruments running POPs analyses always run **CHBr2**. If SIM windows need to be updated, then **CHBr7** should also be run using **CHBrwndw.m**. POPs standards should be run with a conditioning solution corresponding to the samples that will be loaded next on the instrument.

New conditioning solutions are made using SRMs from archived sets that have already been reported. [Section 6.2](#) details how to prepare standards for instrument checks or for sample sets.

### 5.2 Setting Up and Running the Standards Sequences

Standards sequences are already set up and saved on each computer. In ChemStation **Instrument Control**, click **Sequence**, then select **Load Sequence**. The PAHs sequences are named **AlksimCk**, and the POPs sequences are named **CHBrCk**. After loading the appropriate sequence, click **Sequence** then select **Edit Sequence**. The sequence should consist of conditioning runs and the appropriate standards; however, the standard for SIM windows may not always be there, or may be present when not needed. To add the SIM window standard, just copy and paste a standard run as a new run, then rename the standard (both the sample name and the data file name). Ensure that the vial number is correct. In the case of **CHBr7**, the method should be changed to **CHBrwndw.m/CHWNDW**, and for **AHP7** the method should be changed to **AHscan**. To remove an unneeded SIM window standard, simply right-click its row and select **Delete Row**. When the sequence is complete, click **OK**. A message may give an alert that more than one sample has the same data file name because the conditioning runs have the same name. Conditioning run data is typically not used, in which case it is acceptable for new conditioning runs to overwrite previous conditioning runs. Click **OK** to return to **Instrument Control**. Click **Sequence**, then select **Run Sequence**. Here the data path must be renamed. Each standard run is named **AlksimCk** or **CHBrCk** and the date in *MMDDYY* format. For example, if an **AlksimCk** is run on 3 November 2018, the data folder is named **AlksimCk110318**. Make sure the data path starts on Drive D:. All data paths should begin **D:/MSDCHEM/1/DATA/**. After the data path has been set, enter a description of the run along with your (the operator’s) initials. Normally, both of those

will default to the last saved sequence, so only the operator's initials usually need to be changed. If the field for entering the operator's initials is grayed out, click **Full Method** at the top of the window. This should unlock all the fields. Click **OK** (do not run the sequence at this point). Click **Sequence** again and select **Print Sequence**. This printout should be placed next to the instrument, so other users know what the system is working on. Do a last check by selecting **Sequence** and then **Edit Sequence**. Check the data path, the vial numbers, the methods, and the data file names, and fix any errors. Click **OK**. Finally, place the standards and conditioning sample into the sample tray in the correct order. The conditioning sample should be in Position 1, followed by the standard(s). Make sure the autosampler arm is in the ready position, meaning it should not be in the parked position (Instrument 1 has no ready position). If the arm is in the parked position, press the button labeled **P** just below the tray. This will move the arm to the ready position. The standards are ready to run, so click **Sequence**, select **Run Sequence**, and then click **Run**. Everything is automatic and should only need to be briefly checked every so often to make sure the runs are going smoothly.

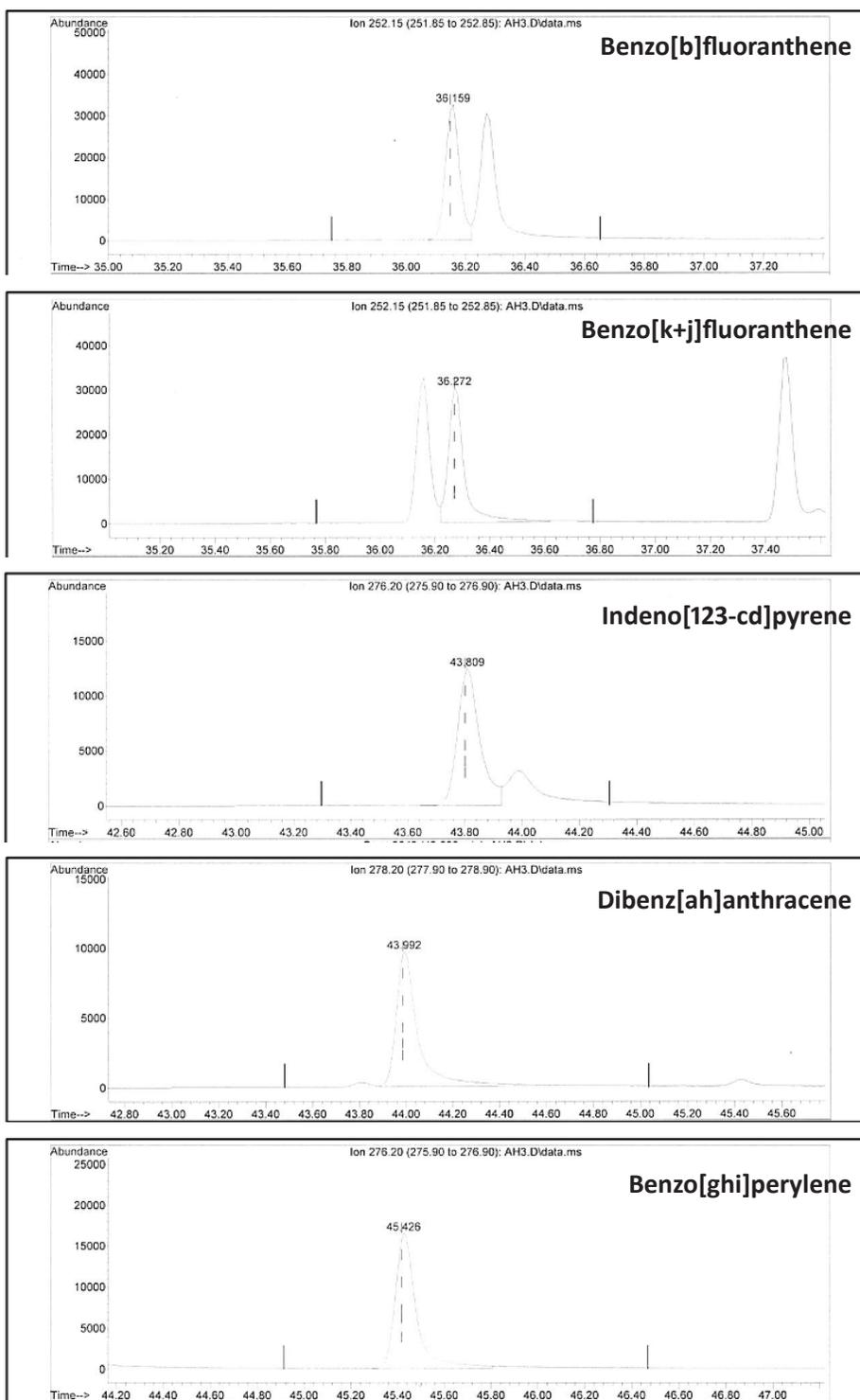
After the sequence is complete, the standards need to be recapped to prevent evaporation. Standards used for checking the instruments are stored on the leftmost side of the sample tray; however, if the lowest-level standard (e.g., **AHP1A** or **CHBr2**) has concentrated in the vial due to evaporation (e.g., if run overnight), then discard the vial.

### 5.3 Checking the Standards

When checking the standards, whether they were run for POPs or PAHs, there are specific analytes to monitor for sensitivity and others for peak shape and resolution.

#### 5.3.1 Standard Used to Check Peak Shape for PAHs Analysis (AHP4A)

**AHP4A** has five compounds: benzo[b]fluoranthene, benzo[k+j]fluoranthene, indeno[123-cd]pyrene, dibenz[ah]anthracene, and benzo[ghi]perylene. They are checked for resolution, peak shape, and tailing (Example 6). Peaks that are close together should have deep valleys between them (resolution). The ends of the peaks should resemble the beginnings. The end of the peak may start trailing off with a longer slope—this is called tailing. Slight tailing is normal, especially in the high-end peaks (indeno[123-cd]pyrene, dibenz[ah]anthracene, and benzo[ghi]perylene); however, significant tailing indicates issues with the system.



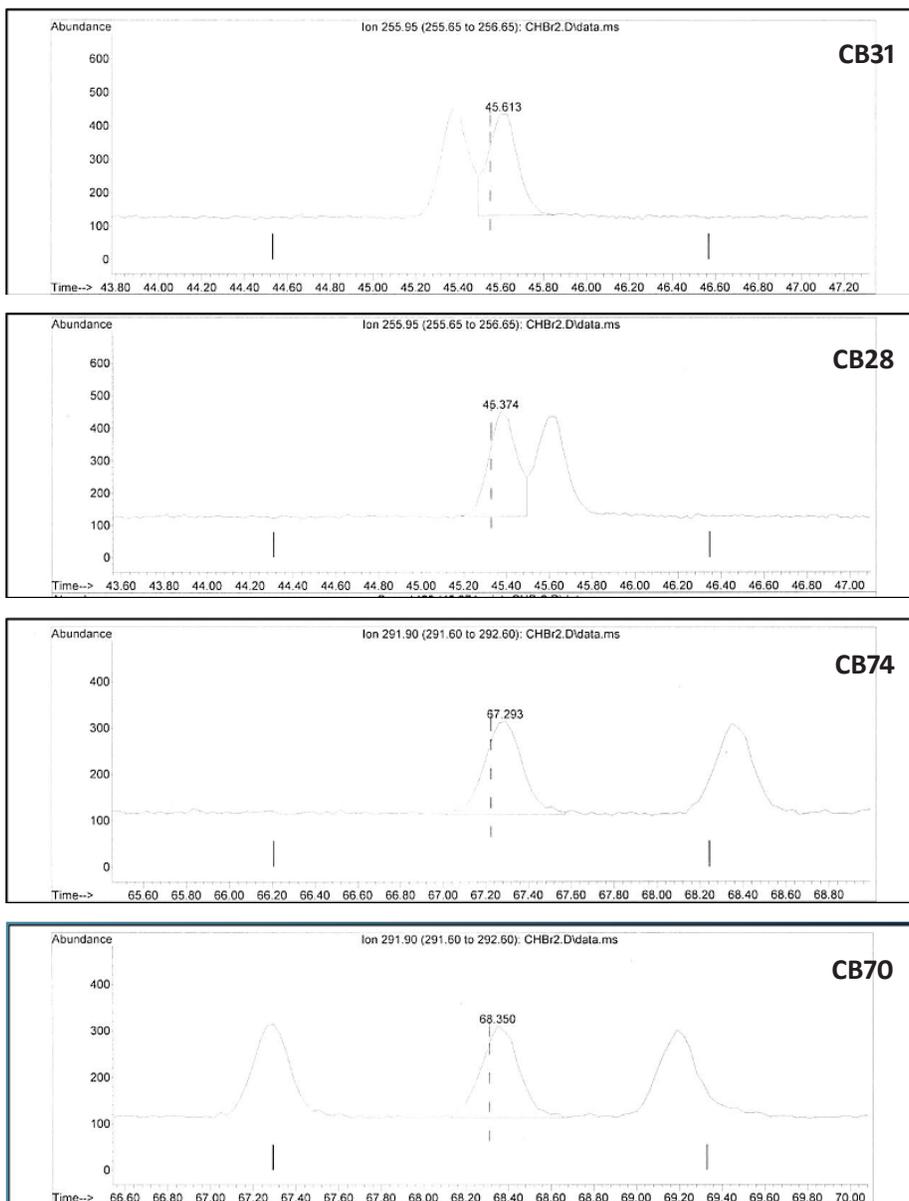
Example 6. Suggested analytes to monitor in **AHP4A** standard, with acceptable peak shapes.

### 5.3.2 Standard Used to Check Sensitivity for PAHs Analysis (AHP1A)

**AHP1A** has five compounds that are checked for sensitivity: hexamethylbenzene, trimethylnaphthalene, indeno[123-cd]pyrene, dibenz[ah]anthracene, and benzo[ghi]perylene. Hexamethylbenzene should have a response of at least 4,000,000. The others should have responses of at least 1,000. Low responses can create tailing and low recoveries in samples.

### 5.3.3 Standard Used to Check Peak Shape and Sensitivity for POPs Analysis (CHBr2)

**CHBr2** has four polychlorinated biphenyl (PCB) compounds (CB31, CB28, CB74, and CB70) that are checked for peak shape, resolution, and tailing (Example 7). Peaks that are close together should have deep valleys between them (resolution). The ends of the peaks should resemble the beginnings. If the peaks are tailing significantly, this may indicate issues with the system.



Example 7. Suggested analytes to monitor in **CHBr2** standard, with acceptable peak shapes.

## 5.4 Updating SIM Window Times

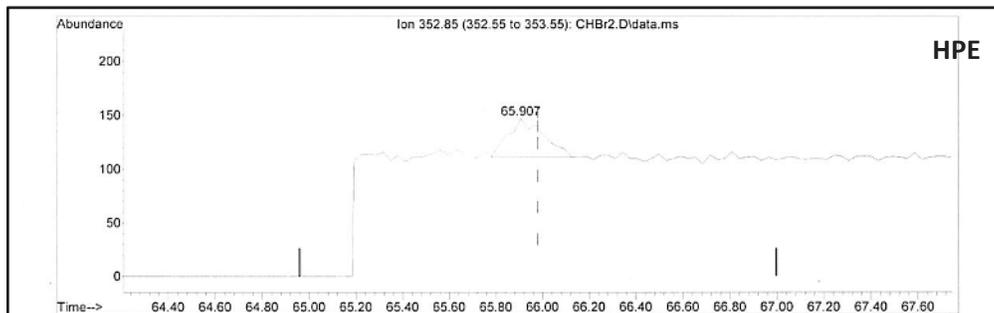
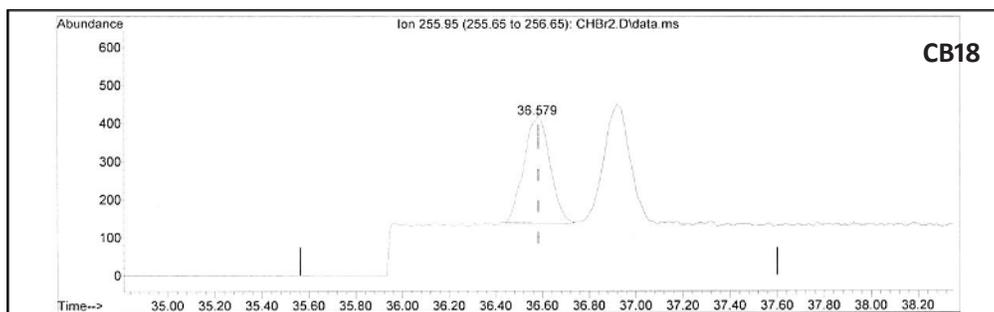
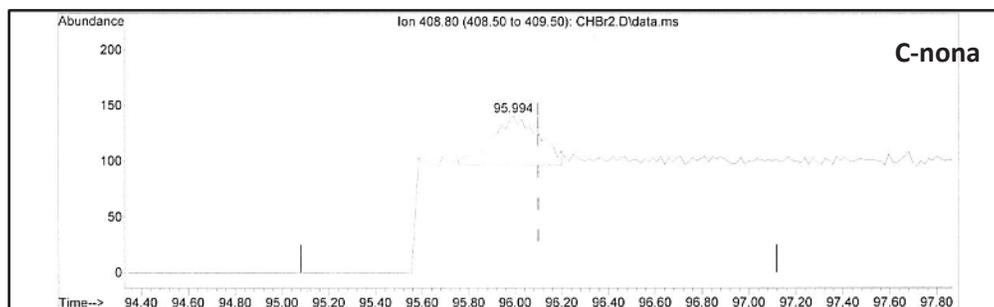
It is essential to keep an eye on how close your peaks are getting to the edge of the SIM window as you run sample sets on the instrument. When the beginning or end of any POPs peak gets within 0.5 min of the SIM window cut off, or when you have conducted major maintenance on the instrument, it is time to update the SIM window times in the methods. For PAHs, you will update the SIM window times every time you do any maintenance on the instrument.

### 5.4.1 PAHs SIM Windows

SIM windows need to be checked on PAHs instruments (e.g., in methods **AHsim.m**, **Alksim.m**, **Alksim2.m**, or **Alksim3.m**) before every set. **AHP4A** is the standard used to calculate PAHs SIM window times—or **AHP7A** run with **AHscan.m** if the retention times have shifted so much that any peaks are not within their windows. Print a report for **AHP4A** or **AHP7A** that has correct retention times. Use the Excel spreadsheet labeled **AHSIM Windows** on the #5 computer. Input the retention times for the indicated analytes, and the new SIM window times will be calculated. Print the Excel spreadsheet (with the date) and use it to update the PAHs method. Load the PAHs method to be used (e.g., **Alksim.m**) in ChemStation **Instrument Control**. Click on the quadrupole icon button, then click **SIM Parameter**. The right side has the SIM windows with their current times. Click on a SIM window (the first window never changes, so always start with Window 2), then input the new time on the left. Click **Modify Ion** to save the change. Do this for all SIM windows. Remember to save each method after exiting the **SIM Parameter** window. After SIM windows are updated, the system is ready to run a sample set.

### 5.4.2 POPs SIM Windows

Typically, the SIM windows for POPs instruments need to be updated only after a major column change (after more than one meter of guard column is removed, after trimming both the guard and analytical columns, or after replacing either column). It is good practice to monitor C-nona, CB18, and HPE routinely to see how close to the edge of the SIM window they are getting (Example 8). If the analytes are less than an inch or so from the edge of the SIM window, the **CHBr7** standard should be run using **CHBrwndw.m** as the method to calculate new POPs SIM window times. Use the Excel spreadsheet labeled **CHBrSIM Windows** on the desktop of the #1, #6, or #7 computers. Input the retention times for the indicated analytes, and the new SIM window times will be calculated. Print the Excel spreadsheet (with the date) and use it to update the POPs methods **CHBrStd.m** and **CHBrSim.m**, just like with PAHs. Remember to save each method. After SIM windows are updated, the system is ready to run a sample set.



Example 8. Suggested analytes to monitor for signs that it is time to update SIM window times in the POPs methods.

## 6 Running a Set of Samples

The information needed to run a set of samples includes: the project, the matrix (blubber, fish muscle tissue, shellfish, water, etc.), the analyte list, the calibration standards needed, the acquisition method(s) to use, and which GC/MS system to use. Most sample sets have an SRM (standard reference material), a method blank, a high-performance liquid chromatography (HPLC) blank, and an internal standard check vial. Some sets will have more than one SRM, a solvent blank, a spiked blank, or a spike vial. Sample sets can consist of 1–13 field samples. The sequence of runs will depend on the number and types of samples and calibration standards.

### 6.1 Updating the Logbook

After loading a sample set on the GC/MS, the specifics of the set should be recorded into the system's logbook. The information about the set, the set name, the standards, and the total amount of injections should be logged within the logbook.

*Example of a logbook entry for running a set of samples:*

- “Running a set of turtle liver for PAHs/alkyls \DH2745. AHP1A–AHP8A. +32.”

### 6.2 Preparing Standards

All sequences are run with a series of multilevel calibration standards. For POPs sets, run the **CHBr** series (**CHBr1–CHBr11**), and for PAHs sets run the **AHPA** series (**AHP1A–AHP8A**). POPs sets may require the high molecular weight polychlorinated biphenyls (**HPCB**) series (**HPCB-1–HPCB-7**) or the **PCB11** series (**CHBrX2-1–CHBrX2-5**). These standards are summarized in Table 1. Sets may not require every standard in the series. The series of standards required for the set to be analyzed will be specified on the folder. Standards should be prepped after a sequence has been written out and the number of reproducibility standards has been determined.

Standards are stored in a freezer (**ECP 21**) in **Lab 253E** and must be brought to room temperature before use. The PAHs standards should be covered with a piece of aluminum foil to protect them from UV during this process. Limited volume insert (LVI) vials with inserts should be labeled while the standards warm to room temperature (Figure 22). When the standards have reached room temperature, mix each standard in its vial using the Vortex Mixer. Use a disposable 5" glass pipette to transfer approximately 100–150  $\mu\text{L}$  of standard (slightly above the neck of the insert) into each respective labeled LVI vial and ensure the caps are firmly closed and that the septa dimple. Mark the levels on each LVI vial so any evaporation can be detected. Each reproducibility standard (**CHBr6** or **AHP4A**) will need at least three LVI vials, depending on the length of the sequence. A reproducibility standard should be run approximately every 12 injections. Longer sequences or double sets will require up to five reproducibility standards.



Figure 22. AHP standard, properly prepared, labeled, level-marked.

Table 1. Standards used to run the POPs and PAHs analyses.

GC/MS Standards			
CHs	Main CH Standards run with every set (levels depend on the samples being run)	Special Case Standards	
		PCB11 Standards (run mostly for PSEMP <sup>†</sup> sets)	HPCB Standards (run with special case marine mammal sets)
	CHBr1	CHBrX2-1	HPCB-1
	CHBr2	CHBrX2-2	HPCB-2
	CHBr3	CHBrX2-3	HPCB-3
	CHBr4	CHBrX2-4	HPCB-4
	CHBr5	CHBrX2-5	HPCB-5
	CHBr6*		HPCB-6
	CHBr7		HPCB-7
	CHBr8		
	CHBr9		
	CHBr10		
	CHBr11		
AHs	AHP Standards		
		AHP1A	
		AHP2A	
		AHP3A	
		AHP4A*	
		AHP5A	
		AHP6A	
		AHP7A	
		AHP8A	

\* Reproducibility standard. † Puget Sound Ecosystem Monitoring Program.

### 6.3 Setting Up a Sequence

The set folder has a white sheet that should indicate the predicted levels of POPs. This will indicate the range of standards to use for the run. For example, a moderate range for POPs may mean only standards **CHBr2** through **CHBr9** need to be used. A high range for PAHs may mean all standards, **AHP1A** through **AHP8A**, need to be run. The folder should have the information required to choose the standards needed. Each set needs reproducibility standards that are used to evaluate the performance of the instrument while running a set, so these standards are repeated at least three times (at the beginning, middle, and end of the sequence). POPs sets use **CHBr6** as the reproducibility standard, and PAHs sets use **AHP4A** as the reproducibility standard (Tables 1 and 2).

The sequence starts with a conditioning run and a reproducibility standard, then incorporates the HPLC and method blanks and the SRM, as follows:

1. Conditioning sample.
2. Reproducibility standard.
3. HPLC blank.
4. Method blank.

Table 2. Standards, acquisition methods, and data processing methods and their corresponding matrix and target analyte lists.

Matrix	Analyte List	Standards (series) <sup>a</sup>	Repro. Standard	Methods to Use				Point-to-Point	Calibration Table <sup>b</sup>
				Acquisition Samples	Acquisition Standards	Repro. Method	Data Processing		
tissue, sediments for individual PAHs only	individual PAHs, no alkyl groups, no BNTs	AHPA	AHP4	AHSIM.m	AHSIM.m	AHPrepro1.m	AHcal.m	AHptpt.bas	AH_2-6.csv
PSEMP tissues, Mussel Watch tissues	individual PAHs and alkyl groups, no BNTs; includes 9MP, 2MP, 3MP, 2MA, DMP	AHPA	AHP4	AlkSIM.m	AlkSIM.m	AHPrepro1.m	AHP1Cala.m	AHP5ptpt.bas	AHP1_27A.csv or AHP1_28A.csv
stormwater, seafood	individual PAHs and alkyl groups, no BNTs	AHPA	AHP4	AlkSIM.m	AlkSIM.m	AHPrepro1.m	AHP1Cal.m	AHP1ptpt.bas	AHP1_2-6.csv or AHP1_2-8.csv
oiled water, WAF (Ecotox water samples)	individual PAHs and alkyl groups, phytane, BNTs	AHPA	AHP4	AlkSIM3.m	AlkSIM3.m	AHPrepro3.m	AHP3Cal.m	AHP3ptpt.bas	AHP3_1-8.csv or AHP3_2-8.csv
tissues, QA exercises, special projects	individual and alkyl PAHs, including 3MP, 9MP, 2MA, BMP, BNTs (no phytane)	AHXC	AHXC4	AlkSIM2.m	AlkSIM2.m	AlkX3rep.m	AlkX3Cal.m	AlkX3pt.bas	AlkX31-6.csv or AlkX31-8.csv
tissues	standard POPs	CHBr	CHBr6	CHBrSim.m	CHBrStd.m	CHBrRepro.m	CHcal.m BRcal.m	CHptpt.bas BRptpt.bas	CH7-2.csv Br7-2.csv
tissues for POPs with PCB-11	POPs including PCB-11	CHBr CHX2	CHBr6	CHBrSim.m	CHBrStd.m	CHBrRepro.m	CHX2cal.m BRcal.m	CHXptpt.bas BRptpt.bas	CHX2.csv Br7-2.csv
tissues (NRDA)	POPs including HMWPCBs 196, 200, 201, 202, and 207	CHBr CHX	CHBr6	CHBrSim.m	CHBrStd.m	CHBrRepro.m	CHXcal.m BRcal.m	CHXptpt.bas BRptpt.bas	CHX11-2.csv BR9-2.csv

<sup>a</sup> The default range of standards for tissues (low) is **CHBr2-7, AHPA2-6**. For tissues (medium/high), use **CHBr2-9, AHPA2-8**. For tissues with high DDTs, use **CHBr2-11**. For tissues with CB11, use **CHBr2-7** and **CHBrX2-1-5**.

<sup>b</sup> Calibration tables are project-dependent and depend on the range of standards run with the set; the range of standards to use will be noted on the cover.

Key: BNT = benzonaphthothiophene, DMP = dimethylphenanthrene, MA = methylanthracene, MP = methylphenanthrene, WAF = water associated fraction.

For NRDA projects, also include **CHBr1** and **CHBrX2-1** for POPs, and **AHPA1** for PAHs.

AHXC MLs contain the additional MPs in the standard; the PSEMP method uses AHPA MLs and the response of 1MP is used to calculate the other MPs. AHXC is a series of multilevel standards that contains extra MPs listed, plus 2-methylphenanthrene and a benzonaphthothiophene, that are not contained in AHP-series multilevel standards.

This file is on Google Drive in **ECP Documents > SOPs > GC/MS SOPs**.

5. SRM.
6. First sample.
7. First multilevel standard.
8. Second sample.
9. Second multilevel standard.

If the set has two SRMs, the second SRM can replace the second sample. The sequence continues to alternate standards and samples. A reproducibility standard should be inserted after approximately every 9–13 runs, and follows a sample or conditioning run. The number of samples/standards between each reproducibility standard should be as even as possible. For example, if 12 runs occur after the first reproducibility standard, then a range of 11–13 runs needs to occur after the second reproducibility standard. Every sequence should end thusly:

29. Sample (or conditioning run).
30. Check vial.
31. Sample (or conditioning run).
32. Reproducibility standard.

Conditioning runs can be used in the place of samples if there are not enough samples remaining to alternate with the standards and check vial. Conditioning runs are just samples run with the conditioning method, and the data collected are not used.

## 6.4 Building a Sequence

It is easiest to open up a system check sequence and clear out the data in all of the rows. Then, starting with a blank sequence template, enter in the information pertaining to the set that will be loaded on the instrument.

### 6.4.1 Sequence Columns

The table columns that the information needs to be entered into are the following: **Sample**, **Vial**, **Method/Keyword**, **Data File**, and **Type/Line**. It is important to enter the information specifically, as a minor detail might cause the system to pause during a run.

#### 6.4.1.1 Sample

Starting with the **Sample** column, type in the sample number and/or the standard information. All the standards should begin with the set number (e.g., **PS3521AHP2A**). Each reproducibility standard should have a letter at the end of the sample name indicating its position in the sequence, such that the first reproducibility standard is *a*, the second is *b*, the third is *c*, and so on (e.g., **PS3521AHP4Aa**). Sample numbers do not require extra information. A sample used as a conditioning run should be labeled with *cond* (e.g., **124-2376cond**). Once all of the required standards and samples have been correctly entered into the **Sample** column, move on to the **Vial** column. If the set requires the first half of the sequence to be rerun at the end of the sequence due to poor reproducibility standards, the corresponding sample names must be copied at the end of the sequence. The sample names should not be altered.

### 6.4.1.2 Vial

The **Vial** column pertains to the vial position in the autosampler tray. Enter **1** for the position of the first reproducibility standard. Highlight the rest of the column below that, right-click, and select **Fill column in increment**. Make sure all conditioning runs and standard reruns, if any, have the appropriate vial position numbers. If the set requires the first half of the sequence to be rerun at the end of the sequence due to poor reproducibility standards, the corresponding vial numbers must be copied at the end of the sequence. New standards should be made up, including another reproducibility standard, so those vials will have new vial position numbers.

### 6.4.1.3 Method/Keyword

Change the method by right-clicking on the cell and selecting **Browse for Method**. A conditioning run uses the **Cond.m** method. All PAHs samples and standards use **Alksim.m**, **Alksim2.m**, or **Alksim3.m**, POPs samples use **CHBrSim.m**, and POPs standards use **CHBrStd** (Table 2). This process can be sped up by right-clicking and copying the corresponding method to the row and pasting it inside of the cell. If the set requires the first half of the sequence to be rerun at the end of the sequence due to poor reproducibility standards, the corresponding method/keyword must be copied at the end of the sequence.

### 6.4.1.4 Data File

Fill the **Data File** column by copying the **Sample** column and pasting it in the cells within the **Data File** column. A data file name can only have eight characters, so follow these rules: Delete the set numbers from all the standards and remove the *A* from the end of the AHP standards. For example, **PS3521AHP3A** would have a data file name of **AHP3**. Remove the *b* from reproducibility standard *b* for sets run for both PAHs and POPs. Remove sample numbers from conditioning runs, so the data file for **124-2376cond** would read **cond**. If the set requires the first half of the sequence to be rerun at the end of the sequence due to poor reproducibility standards, the corresponding data file names of the samples/standards that are being rerun must be copied at the end of the sequence and an *r* added at the end. Because data files can only have eight characters, you might have to shorten the sample data files. For example, **124-2375r** would have a data file of **-2375r**.

## 6.4.2 Finishing and Saving the New Sequence

When you are done creating the list of runs in the new sequence, click **OK** at the bottom of the page. If there are several conditioning runs, a warning will pop up about data files with the same name; click **Yes** to continue. Once the sequence has been created, it needs to be saved as a specific file. Click **Sequence**, select **Save Sequence As...**, and name it the same as the set number. Make sure to include information about what the set is being run for, and/or how many times it has been run, and if it has been rerun through HPLC (e.g., **PS3521Ahr**). An *rh* after the set name signifies that the set was rerun through HPLC, whereas an *r* after the set signifies that the set is being rerun on the GC/MS a second time (there might be sets with *r2* or *r3* depending on how many times it takes to pass QA).

Now that the sequence of runs has been generated, the final steps are to edit the sequence information and save the sequence with the appropriate name. Click **Sequence** at the top of the screen and select **Run Sequence**. For **Method Sections to Run**, select **Full Method**. For **Sequence Barcode Options**, select **Disable Barcodes for this Sequence**. Edit the **Sequence Comment** to match the title of the set, found on the front of the folder (e.g., **PSEMP 2017 Herring Eggs**). The **Operator Name** should be the initials of the operator. For the **Data File Directory**, ensure the data path is correct, starting from the D: drive and adding the set name for the data folder where the set's data will be stored (e.g., **D:\MSDCHEM\1\Data\PS3521Ahr**). It is important that the set's data folder name matches the set name at the end of the sequence. For **Pre-seq Macros/Commands** and **Post-seq Macros/Commands**, the fields are left blank. Click **OK** when finished. Make sure you save. Print out a copy of the sequence and place it next to the instrument (Examples 9 and 10).

```

Sequence Name: D:\MSDCHEM\1\Sequence\M3609AH.8
Comment: Otter scat 2016
Operator: GRB
Data Path: D:\MSDCHEM\1\DATA\M3609AH\
Instrument Control Pre-Seq Cmd:
Data Analysis Pre-Seq Cmd:
Instrument Control Post-Seq Cmd:
Data Analysis Post-Seq Cmd:

Method Sections To Run      Sequence Barcode Options
(X) Full Method             ( ) On Mismatch, Inject Anyway
( ) Reprocessing Only      ( ) On Mismatch, Don't Inject
(X) Barcode Disabled

-----
Line      Sample Name/Misc Info
-----
1) Sample 5 condn CONDN condn
2) Sample 1 AH94Aa ALKSTM M3609AH94Aa
3) Sample 2 140-1642 ALKSTM 140-1642
4) Sample 3 140-1639 ALKSTM 140-1639
5) Sample 4 140-1641 ALKSTM 140-1641
6) Sample 5 140-1626 ALKSTM 140-1626
7) Sample 6 AH22 ALKSTM M3609AH22A
8) Sample 7 140-1627 ALKSTM 140-1627
9) Sample 8 AH23 ALKSTM M3609AH23A
10) Sample 9 140-1628 ALKSTM 140-1628
11) Sample 10 AH25 ALKSTM M3609AH25A
12) Sample 11 140-1629 ALKSTM 140-1629
13) Sample 12 AH94 ALKSTM M3609AH94Ab
14) Sample 13 140-1630 ALKSTM 140-1630
15) Sample 14 AH95 ALKSTM M3609AH95A
16) Sample 15 140-1631 ALKSTM 140-1631
17) Sample 16 AH97 ALKSTM M3609AH97A
18) Sample 17 140-1632 ALKSTM 140-1632
19) Sample 18 140-1633 ALKSTM 140-1633
20) Sample 19 140-1634 ALKSTM 140-1634
21) Sample 20 140-1635 ALKSTM 140-1635
22) Sample 21 140-1636 ALKSTM 140-1636
23) Sample 22 140-1637 ALKSTM 140-1637
24) Sample 23 140-1644 ALKSTM 140-1644
25) Sample 24 140-1638 ALKSTM 140-1638
26) Sample 25 AH94Ac ALKSTM M3609AH94Ac

Last Modified: Thu Mar 22 14:26:23 2018 Page: 1

```

Example 9. A printout of a sequence created to run a set of samples for PAHs analysis.

```

Sequence Name: D:\msdchem\1\sequence\OH2754.s
Comment: Arkoosh/EPA Salmon for PBDES
Operator: RP
Data Path: D:\MSDCHEM\1\DATA\OH2754\
Instrument Control Pre-Seq Cmd:
Data Analysis Pre-Seq Cmd:
Instrument Control Post-Seq Cmd:
Data Analysis Post-Seq Cmd:

Method Sections To Run      On A Barcode Mismatch
(X) Full Method             (X) Inject Anyway
( ) Reprocessing Only      ( ) Don't Inject

-----
Line      Sample Name/Misc Info
-----
1) Sample 5 CONDN CONDN 128-0586cond
2) Sample 1 CHBR6A CHBRSTD OH2754CHBr6a
3) Sample 2 128-0601 CHBRSIM 128-0601
4) Sample 3 128-0599 CHBRSIM 128-0599
5) Sample 4 CHBR2 CHBRSTD OH2754CHBr2
6) Sample 5 128-0586 CHBRSIM 128-0586
7) Sample 6 128-0600 CHBRSIM 128-0600
8) Sample 7 128-0587 CHBRSIM 128-0587
9) Sample 8 CHBR3 CHBRSTD OH2754CHBr3
10) Sample 9 128-0588 CHBRSIM 128-0588
11) Sample 10 CHBR4 CHBRSTD OH2754CHBr4
12) Sample 11 128-0589 CHBRSIM 128-0589
13) Sample 12 CHBR5 CHBRSTD OH2754CHBr5
14) Sample 13 128-0590 CHBRSIM 128-0590
15) Sample 14 CHBR6 CHBRSTD OH2754CHBr6b
16) Sample 15 128-0591 CHBRSIM 128-0591
17) Sample 16 CHBR7 CHBRSTD OH2754CHBr7
18) Sample 17 128-0592 CHBRSIM 128-0592
19) Sample 18 CHBR8 CHBRSTD OH2754CHBr8
20) Sample 19 128-0593 CHBRSIM 128-0593
21) Sample 20 CHBR9 CHBRSTD OH2754CHBr9
22) Sample 21 128-0594 CHBRSIM 128-0594
23) Sample 22 128-0595 CHBRSIM 128-0595
24) Sample 23 128-0596 CHBRSIM 128-0596
25) Sample 24 128-0597 CHBRSIM 128-0597
26) Sample 25 128-0602 CHBRSTD 128-0602
27) Sample 26 128-0598 CHBRSIM 128-0598
28) Sample 27 CHBR6C CHBRSTD OH2754CHBr6c

```

Example 10. A printout of a sequence created to run a set of samples for POPs analysis.

## 6.5 Running the Sequence

Once the sequence has been generated, print it by going to **Sequence**, and then to **Print Sequence**. Use the printed copy of the sequence to correctly place the samples and standards into the sample tray. Double check that the sample levels are all at the marked lines. Put the autosampler arm into the ready position by pressing the illuminated **P** button found on the front of the autosampler tray. When ready, click **Sequence**, select **Run Sequence**, then click **Run**. The sequence is now automated and will run according to the entries in the sequence. The printed sequence should be left next to the instrument so that, while recapping, samples can be placed back into their correct positions on the tray.

The set of samples is now running on the GC automatically, and the computer is collecting data from the MS. The total run time can be estimated by using these guidelines:

- One POPs sample takes about 2.6 hours to complete.
- One POPs standard takes about 2.5 hours to complete.
- Conditioning runs take about 1 hour to complete.
- One PAHs sample takes about 1 hour to complete.
- One PAHs standard takes about 1 hour to complete.

Thus, if a POPs set has 11 standards (including the reproducibility standards), 17 samples, and four conditioning runs, then the whole set will take approximately 75.7 hours, or approximately three and a half days to complete. The GC/MS system should be checked periodically throughout this time, if possible, to be sure it's running properly.

Samples need to be recapped after their caps have been perforated. Recapping prevents any evaporation. Generally, samples are recapped first thing in the morning and just before the end of the day. Make sure the caps are tight enough to make the septa dimple. Standards that have been injected are removed from the sample tray and placed onto a vial board for disposal. Standards that have not yet been injected should have their levels checked. Standards that have evaporated need to be replaced. Sample sets should not be run over weekends that are longer than two days without being recapped, as they will fully evaporate after several days of being perforated.

Rinse vials should be checked periodically as the sample set runs (see [Section 2.4](#)).

## 6.6 Calculating the Reproducibility Standards

The data files need to be checked on samples and standards that have been analyzed, and the reproducibility standards need to be calculated. Open ChemStation **Data Analysis** and click **Method** at the top of the page, then select **Load Method**. Select the reproducibility processing method that pertains to the set that you are running, either **CHBrRepro** or the appropriate method for PAHs (e.g., **AHPrepr1**, **AHPrepr2**, **AHPrepr3**, or other) depending on what analytes the set was run for. Open the data folder by clicking **File** at the top of the screen and selecting **Load Data File**. Click **Change Path**, search for the correct sample set

folder under **Data**, and click **OK**. Once the folder is open, select the data file you want to open up and look at. When calculating the reproducibility standards, typically the *b* run of either the **AHP4A** or **CHBr6** should be opened first. Quantitate by going up to the top of the screen, clicking **Quantitate**, and then selecting **Calculate**. After the standard has been quantitated, click **View** and select **QEdit Quant Results**. Scroll through all of the analytes and make sure that the right peak is being integrated (use the black binder to assist with retention times and peak shapes if needed). Once all of the correct peaks have been integrated, click **Exit** and save the changes in the next window that pops up.

Now the reproducibility method can be calibrated. Go to the top of the screen and click **Calibrate**, then **Update**. Select **Update One Level**, then click **OK**. Click **No** when asked **Requantitate now?** Select **Update Level**. Click on the squares next to **Retention Time** and **Responses**, as well as the two bubbles next to **Replace**. Click on the level number in the **Existing Level ID** box. Finally, click **Do Update**. Now all of the correct retention times have been saved in the correct reproducibility method. Next, all of the amounts in the reproducibility method must be set to 100%. Click **Quantitate** and select **Calculate**. The **epatemp.txt** file generated and displayed on the screen will show all of the analytes, their retention times and responses, and the amounts set to 100%. Occasionally some analyte amounts will not get set to 100% at this point, in which case all of the above steps in this paragraph should be repeated until all of the responses are set to 100%. Save the method again. Right-click and select **Print** to print the report. A printed copy of reproducibility *b* with the responses set to 100% should be placed in the sample set's folder.

With the reproducibility method still open in **Data Analysis**, open up the next reproducibility standard (reproducibility standard *a*). Click **Quantitate** and select **Calculate**. The **epatemp.txt** file generated and displayed on the screen will show all of the analytes and their retention times and responses in that standard, as well as the calculated percentage amounts based on reproducibility standard *b* having been set to 100%. Right-click and print the report for the set's folder. All of the reproducibility standards should be calculated in this way, and their reports printed for the set's folder. Monitor the amounts to make sure that they don't decrease or increase drastically as the set runs. The relative standard deviations (RSDs) for POPs and PAHs have to be 15% or less, and the RSDs for PBDEs have to be 20% or less for them to pass QA. The RSD calculator that is located on the #6 computer can be used to calculate the RSDs of a set to monitor how the set is running, whether the set needs to be rerun, or whether the first half of the sequence should be rerun at the end of the sequence.

## 6.7 Restarting a Sequence After an Error or the Run Has Been Aborted

If an error occurs mid-run, or the sequence needs to be aborted for one reason or another, there is an option to restart the sequence at a certain position in the sequence. Begin by going to **Sequence** at the top of the screen and reload the current sequence (save any changes to the sequence, if prompted). Click **Sequence** again, and select **Position and Run Sequence**. The sequence that was last loaded should pop up. Scroll down and find the line that the sequence should be restarted on, highlight the line, and click **Start**. All of the data from the first part of the run and all of the data acquired in the section of the sequence that was restarted will be in the folder they were initially assigned to.

## 6.8 Unloading the Autosampler Tray After the Run

After the sequence has finished, all of the samples should be removed from the autosampler tray, recapped, placed back on the labeled board, and placed in the refrigerator in **Room 317E** until the data have been reported out. When recapping the samples, make sure that the caps are on tight enough that the septa dimple. It is also a good idea to check the volumes of the samples, assessing whether they need a drop or two of isooctane to bring them back up to the volume marked by the extractionist. The standards can be placed on the discard tray and, eventually, properly discarded.

## 7 MS Maintenance

MS parts that require maintenance include the analyzer, the rough pump, and the calibration gas. The MS analyzer primarily consists of the ion source, the mass filter (quadrupoles), and the electron multiplier (EM). A clean ion source may need to be installed periodically ([Section 7.3](#)), and the EM may need to be replaced infrequently ([Section 7.4](#)). The mass filter typically will not need any maintenance. The rough pump oil needs to be replaced yearly ([Section 7.6](#)), and the calibration gas needs to be refilled as needed ([Section 7.7](#)).

Before performing MS maintenance, the MS must be cooled and fully vented to ambient pressure ([Section 7.1](#)). After completing maintenance in the MS, the MS must be pumped down to be under vacuum before it can be used ([Section 7.2](#)). The toolkit holding the MS tools can be found in the desk under GC/MS #5. These tools should only be used when performing maintenance or repairs on analyzer components. The radio frequency voltage might need to be adjusted after any MS maintenance ([Section 7.5](#)).

### 7.1 Venting the MS

In ChemStation **Instrument Control**, click **View**, select **Tune and Vacuum Control**, then click **Vacuum**. Select **Vent** and click **OK**. This will turn off the turbo pump and start cooling the MS, generally taking about an hour to complete. It will also automatically shut the auxiliary heater and the oven off, so you should manually reset the oven temperature to 30°C. Once the MS is cool and the turbo pump has stopped, **Instrument Control** will shut itself down. When the system is cool, turn the MS off by pressing the power button on the bottom of the instrument. Turn off the rough pump, either by unplugging the pump or by using the power switch. Open the cover in front of the source by pushing down on the top of the cover and pulling it out, then vent the system using the vent valve knob on the front of the MS ([Figure 23](#)). Wait until the hissing stops, then close the valve.



Figure 23. Vent valve knob. Also shown is the calibration gas bulb, filled with PFTBA (perfluorotributylamine).

### 7.2 Pumping Down the MS

Once the MS has been closed, it can be pumped down. Turn on the rough pump and push on the panel for a few seconds to ensure a tight seal. Latch the panel closed and replace the cover panel. Push the MS power switch on, then the GC power switch on, then the computer power switch on. Restart ChemStation **Instrument Control** on the computer (this will take a few minutes). Click **OK** on the temperature window that pops up. Once the program is started, click **Abort** then click **OK**. Click **View**, then select **Tune and Vacuum Control**. Click **OK** again when the temperature window pops up. Click **Vacuum**, then select **Pump Down**. Click **OK** on all the windows that pop up. The **MS Source Target** temperature should be 300°C and the **MS Quad Target** temperature should be 150°C. **Instrument Control** should reheat the ovens, but check the GC control panel to make sure. The system needs to pump down for a minimum of 4 hours before the air/water spectrum is checked ([Section 4.2](#)), but for best results it is ideal to allow the system to pump down overnight before running any tests.

## 7.3 The Ion Source

The ion source ionizes compounds so they can be analyzed. The source is housed in the MS at the front of the analyzer and can be seen through the clear panel (Figure 24). A clean ion source needs to be installed periodically. Continued use of the MS will make the source dirty, and a dirty ion source will create problems in the data (see [Section 8.3](#)). It is also possible that both filaments burn out before the source is dirty, which also requires replacement of the source. Before removing the ion source from the MS, the MS must be cooled and vented to ambient temperature and pressure. After the ion source is replaced in the MS, the MS must be pumped down to under vacuum before the MS can be used.

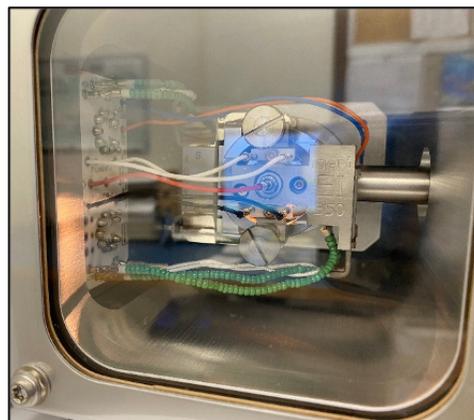


Figure 24. View of ion source through the MS window.

### 7.3.1 Updating the Logbook

When an ion source is removed and a new source is installed, it is important to log the information into the system's logbook. It is good practice to keep track of how long an ion source has been installed for, when troubleshooting issues down the line. It is advised to highlight this entry so it can be easily found.

*Example of a logbook entry for installing a new ion source:*

- “Cooled GC & MS. Shut down vacuum pumps and vented the system to atmosphere. Opened the analyzer and installed a clean ion source; inspected analyzer and phalange O-ring for any visible problems; everything normal. Closed analyzer, allowing system to pump down/up-heat/equilibrate overnight.”

### 7.3.2 Removing the Source

In order to remove the source after the system has been properly vented, open the analyzer chamber by using the top latch to open the side chamber. Never touch the ion source or panel with bare hands; clean gloves must always be worn while working inside the MS. Next, loosen the two large screws that hold the panel in place at the bottom left and the top right; the screws may require a flathead screwdriver. Unplug the two electrical leads at the front of the panel, and push the cords down so they will be clear of the panel. Swing the panel open. There are several electrical leads running to and from the ion source that need to be disconnected. Disconnect the black, white, and red leads from the front of the source and the blue and orange leads from the right side of the source. The white and green leads on the bottom of the source disconnect from the circuit board and remain connected to the source. The white and green leads at the top of the circuit board running to the top of the quadrupole housing are not connected to the source so they do not need to be disconnected (Figure 25). Use forceps to disconnect the leads, if necessary. Once the leads are disconnected, the screws

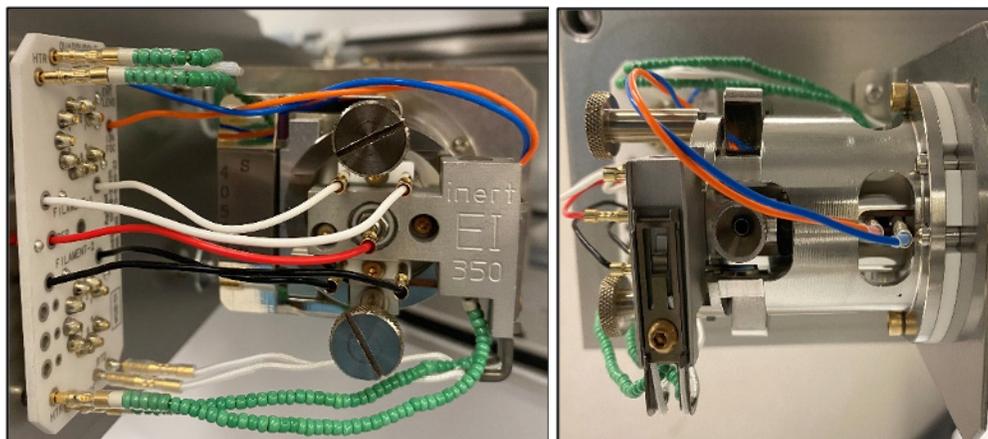


Figure 25. Top view (left) and side view (right) of the ion source inside the MS.

holding the source in place may be removed, using a flathead screwdriver if needed. The source should now pull smoothly out of the MS. Take care not to bend or break the gold contacts that are exposed at the ends of the leads. Place the dirty source back in its plastic case to be stored for later cleaning ([Section 7.3.4](#)). Place a piece of time tape on the top of the case that states that the source needs to be cleaned, the date it was removed, and the instrument it came out of. Refer to [Section 7.3.3](#) to install a new clean source into the MS.

### 7.3.3 Replacing the Source

A clean source is stored in a plastic case, secured on a brace with screws, kept under the GC/MS #1 system. Using clean gloves, remove the screws and carefully lift the source off the brace. Use the small screwdriver to check that the setting screws are tight. Place the clean source into the MS, reversing the steps to remove the source found in [Section 7.3.2](#). The leads may now be replaced. From the circuit board, the blue lead goes in the back on the side of the source, and the orange in front of the blue. The two white leads go on top on the front of the source, the red in the middle, and the two black on the bottom; from the bottom of the source, the two white (braided) leads go into the back part of the circuit board, and the green leads go toward the front of the circuit board.

Before closing up the MS, a few things should be inspected. Check the MS for scorch marks that would indicate a short in the system. Use a flashlight to inspect the seal for any cracking or chipping. A damaged seal needs to be replaced. Run a gloved finger along the black ring-seal, pushing in any bumps, to ensure it is properly seated. After a successful inspection, the analyzer chamber can be closed. Push the panel back into place, then tighten the two screws holding it. Reattach the two electrical leads. The MS can now be pumped down. Proceed to [Section 7.2](#).

### 7.3.4 Cleaning the Source

Dirty ion sources can be cleaned and filaments refurbished and used again. The electron ionization (EI) sources, both dirty and clean, are stored in plastic cases with mounting hardware specifically for the source (Figure 26). The source is held securely in the case by two screws, and it is kept in the desk cabinet under the GC/MS #1 system.

Gather the black folder of tools (Figure 27), along with two new filaments and the green micro-abrasive paper. Place a clean piece of aluminum foil down as your working surface and put on clean white gloves or new nitrile gloves. Remove the dirty source from the protective plastic case and stand it up using the stainless-steel block stand. Designate two beakers, one for parts of the source that will not be cleaned or sonicated (Beaker A), and one for parts that will be sonicated (Beaker B). Start disassembly by unscrewing the two small screws holding the filaments in place on either side of the source. The used filaments can be disposed of, and the screws can be placed into Beaker B. Using the black-handled socket wrench, loosen the repeller nut and place it in Beaker B. Place the metal washers in Beaker B. Remove both repeller insulators and examine them. If they appear worn, cracked, or dirty, replace them with new ones. If the insulators look to be in good shape, place them in Beaker A, as they do not need to be sonicated. Place the repeller aside in order to be cleaned with alumina. Now, take the source heater assembly and place it in Beaker A, cover the beaker with foil, and set it aside, away from the area where other parts will be cleaned with alumina. Nothing needs to be done with the source heater unless you need to check resistance. The source heater should measure approximately 20 ohms and the sensor should measure approximately 109 ohms. Using the black-and-red star screwdriver, remove the small gold setting screw from the bottom end of the source body and place it in Beaker B. Unscrew the transfer line socket with a wrench and set it aside to be cleaned. Remove the draw-out plate, extractor, and lens assembly from the source body. The draw-out plate and extractor can be set aside to be cleaned with alumina. Disassemble the lens assembly; the lens insulator can be placed in Beaker B, and both the ion focus lens and the entrance lens can be set aside to be cleaned with alumina. See Figure 28 for the disassembled ion source.

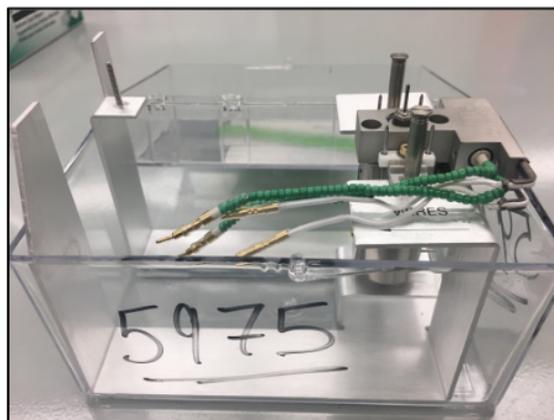


Figure 26. Clean ion source secured in its protective case.



Figure 27. Source cleaning tools.

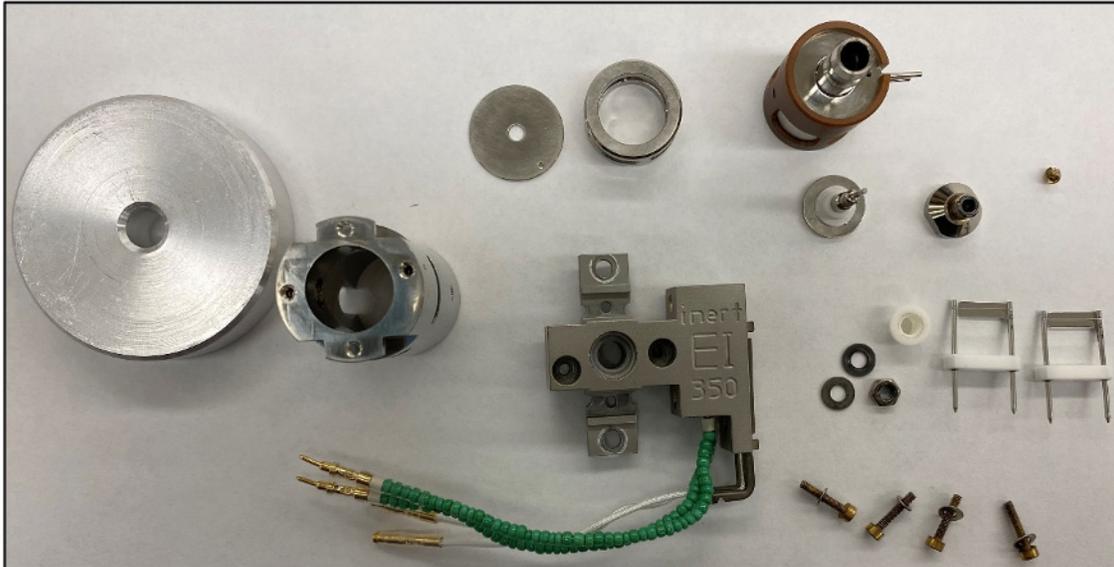


Figure 28. Disassembled ion source.

Before beginning the next step, ensure that Beaker A is covered and placed far away from your working surface. It is also advisable to make sure your tool kit is closed and far away from your working surface as well. Place approximately a teaspoon of the fine grit alumina in a watch glass and add three times that amount of deionized water (DI water) to make a slurry. You will need a pack of small cotton-tipped applicators for cleaning. The alumina and applicators are kept together in the cabinet above the sink. Lay down a clean, lint-free cloth and place a small amount of the slurry on it. It is important to remember to thoroughly clean every surface, hole, slot, and edge, inside and outside, of each of the following parts.

- **The Repeller.** The repeller face (Figure 29) is the most important part of the source to clean. The face of the repeller must remain flat while cleaning. Do not work in circles, as this will wear down the edges. Press it down flat to the bench in the slurry on the lint-free cloth, then move it back and forth 4–5 times. Turn it 90° and repeat the process until it is completely clean, applying only light pressure. The dark color that comes off is not a sign of a dirty source; rather, it is metal that has been removed. Deposits on the face are already gone before you see this. Clean the back using cotton-tipped applicators and slurry. Clean the edge by spinning it on the lint-free cloth in the slurry paste. Once the repeller is thoroughly cleaned, rinse it with DI water over a beaker and place it in Beaker B.
- **The Source Body.** Using cotton-tipped applicators and slurry paste, clean the inside and outside of the source body (Figure 30), including the slots, paying attention to the areas mentioned above. Clean out the electron beam hole last, using the wooden



Figure 29. Repeller.



Figure 30. Ion source body.

end of the cotton-tipped applicator dipped into the alumina slurry. There will be burn marks in the spot where the filaments sit; use the green micro-abrasive paper to get rid of most of them, and follow with cotton-tipped applicators and alumina slurry. *Note:* This is the only place where the green micro-abrasive paper should be used while cleaning the source. It is not necessary to completely clean this area as it is not required for ultimate performance. Scrub the area where the draw-out plate sits inside of the body, as this area is a potential source of contamination. Once the source body is thoroughly cleaned, rinse it with DI water over a beaker and place it in Beaker B.

- **Draw-Out Cylinder and Plate.** Both sides of the draw-out plate (Figure 31) should be cleaned with a cotton-tipped applicator and slurry, making sure to clean the inner edge as well. Ensure the face of the draw-out cylinder (Figure 31) is thoroughly cleaned, as well as the lip where the draw-out plate sits. Both parts can be cleaned as was the repeller face: Lay them flat on a lint-free cloth in the alumina slurry. Place a gloved finger over the top and with light pressure, run the parts back and forth 4–5 times, turn 90°, and repeat until clean, flipping them over to do the other side. When they are thoroughly clean, rinse with DI water over a beaker and place them in Beaker B.
- **Ion Focus Lens.** The ion focus lens (Figure 32) should have the entire inside of the cup, the edge of the center hole, and the backside face cleaned with a cotton-tipped applicator and alumina slurry. Make sure to not round off the edge of the center hole while cleaning it. When it has been thoroughly cleaned, rinse with DI water over a beaker and place it in Beaker B.
- **Entrance Lens.** The flat face, the center tube, and the outside end of the entrance lens (Figure 33) should be cleaned with a cotton-tipped applicator or a lint-free cloth, and alumina slurry. The face of the entrance lens can be cleaned by laying it flat on a lint-free cloth in alumina slurry. Using a gloved hand, run it back and forth 4–5 times, then rotate 90° and repeat until the face is clean. Do not work in circles as this will wear down the edges. Then use a cotton-tipped applicator and alumina slurry to clean the rest of the lens. When it has been thoroughly cleaned, rinse with DI water over a beaker and place it in Beaker B.
- **Transfer Line Socket.** Clean the inside of the socket (Figure 34), along with the threaded end, using a cotton-tipped applicator and alumina slurry. Once the socket has been thoroughly cleaned, rinse it with DI water over a beaker and place it in Beaker B.



Figure 31. Draw-out cylinder (left) and plate (right).



Figure 32. Ion focus lens.



Figure 33. Entrance lens.



Figure 34. Transfer line socket.

Once all parts have been cleaned and placed in Beaker B, place a small piece of foil over the top of the beaker and take it to **Lab 253E** to be sonicated. Fill a new, clean beaker with hot water and Micro-90 soap. Using forceps, remove each source part, place it in the new beaker, and let sit for approximately five minutes. Remove each part using forceps and rinse thoroughly under running water until you no longer see soap bubbles. Using forceps, place all of the parts into the solid sonication tray and add DI water until all of the parts are submerged. Ensure there is 1,650 mL of water in the sonicator basin, then place the tray inside and sonicate for three minutes. After the sonication is complete, drain the DI water from the solid tray by pouring it carefully down the sink, making sure no parts fall out of the tray. Take the tray with parts to the fume hood where the sonicator is located.

The next steps will require working in a fume hood while wearing the appropriate PPE, which includes nitrile gloves and protective eyewear. It is also important not to allow the parts to fully dry between sonicating solvents. Start by ensuring there are exactly 1,650 mL of water in the basin of the sonicator. Next carefully pour methanol over the parts in the solid tray until they are all fully submerged. Place the tray into the sonicator and sonicate for three minutes. Drain the methanol after sonication and dispose of it in the appropriate waste container located inside of the fume hood. Now, carefully pour acetone over the parts until they are submerged, place the tray back in the sonicator, and sonicate for three minutes. Drain the acetone after sonicating and dispose of it in the appropriate waste container located inside of the fume hood. Finally, carefully pour methylene chloride over the parts in the tray, place the tray back in the sonicator, and sonicate for five minutes. Drain the methylene chloride and dispose of it in the appropriate waste container inside of the fume hood. Leaving them inside the tray, allow all of the parts to dry and the solvent to evaporate in the fume hood for at least 30 minutes. Do not bake out the parts in an oven to accelerate the drying process. Once the parts are fully dry, remove the clean parts from the sonicator tray using forceps and place them into a clean beaker to take back up to **Room 317E** to assemble. Follow the manual to reassemble the source, making sure to put new filaments on the source heater assembly (Figures 35–38). Once it is reassembled, it can be placed back in its plastic case with labeled time tape on the outside indicating that it is clean and the date it was cleaned.

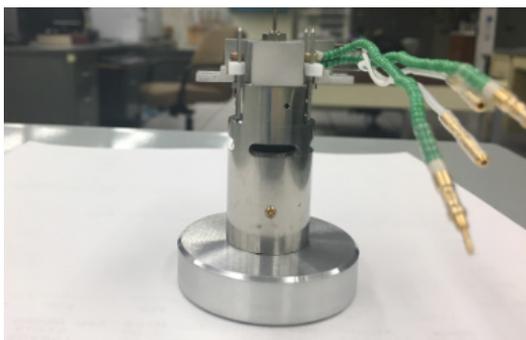


Figure 35. Side view of an assembled EI ion source.



Figure 36. Top view of an assembled EI ion source.

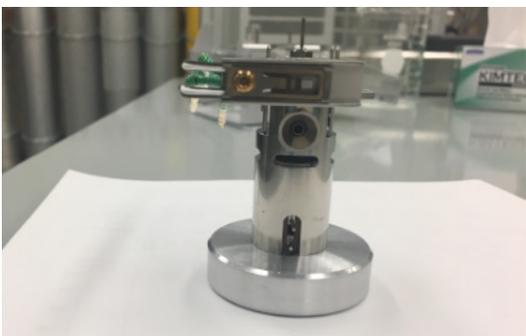


Figure 37. Side view of an assembled EI ion source.



Figure 38. Side view of an assembled EI ion source.

## 7.4 The Electron Multiplier (EM)

The electron multiplier (Figure 39) is on the analyzer panel behind the quadrupoles. A good indicator that the electron multiplier will need to be replaced soon is when, in your **CHtune/sAHtune**, your EM volts are creeping up to 3,000 and your 69 abundance isn't changing significantly. New electron multipliers should only be ordered when you are ready to replace them, because they have short shelf lives.

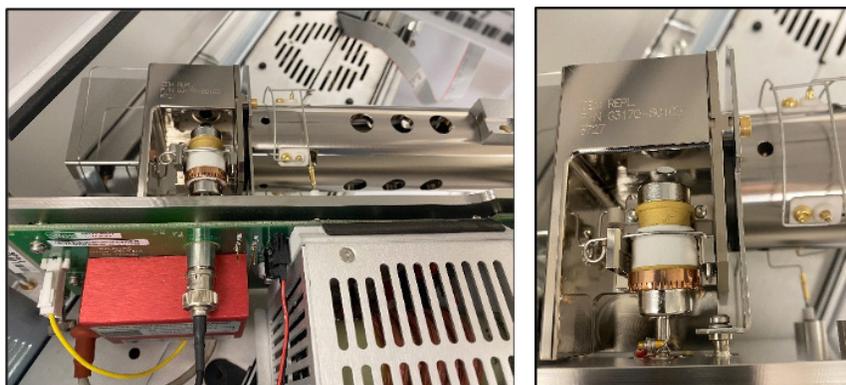


Figure 39. (left) Electron multiplier inside of the analyzer chamber. (right) Zoomed-in image of the electron multiplier.

### 7.4.1 Updating the Logbook

When it is time to install a new electron multiplier, the information should be recorded in the system's logbook. It is good to keep track of how long an electron multiplier has been installed, in case you need to troubleshoot issues down the line. It is advised to highlight this entry so it can easily be found months later.

*Example of a logbook entry for installing a new electron multiplier:*

- "Cooled GC/MS. Shut down vacuum pumps and vented the system to atmosphere. Installed a new EM horn and clean ion source. Allowing system to pump down/up-heat/equilibrate overnight."

## 7.4.2 Removing the MS Analyzer

After the MS has been cooled down, vented, and the entire system powered off, the MS analyzer can be removed. Open the analyzer chamber by opening the side panel using the top latch. Unscrew completely the two large screws that hold the panel in place, one at the bottom left and one at the top right. The screws may require a flathead screwdriver. Unplug the source power cable (dark gray) and the sideboard control ribbon cable (light gray), and push the cords down so they clear the panel door (Figure 40).



Figure 40. (left) Side image of the dark gray source power cable and the light gray control ribbon cable. Both cables need to be disconnected before the panel door can be opened. (right) Front image of the source power cable and the sideboard ribbon control cable.

Swing the panel open (Figure 41). If it has been a while, you might need to use a flathead screwdriver to gently pry it open.

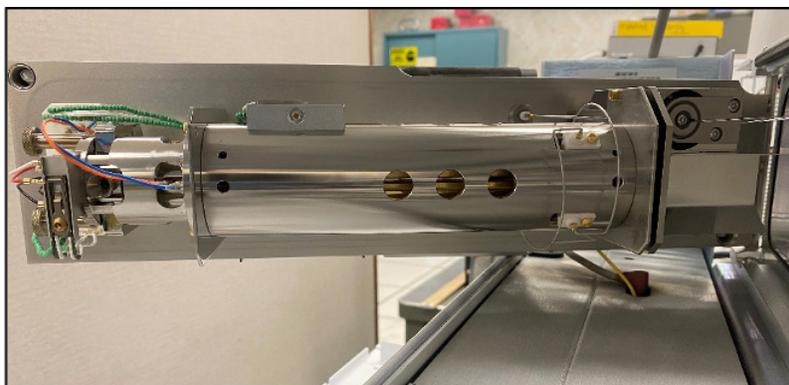


Figure 41. Image of the panel door, open.

Disconnect the high-energy dynode (HED) control cable (yellow), the signal cable (black), and the HED power cable (white with orange tips; Figure 42). At this point, clean white gloves must be worn at all times and no parts should be touched with bare hands. Begin by lifting up on the door and pulling out the analyzer. Set the analyzer down on its own rubber feet (make sure to grab *only* the radiator, *not* the parts with connectors). Place the analyzer on a large piece of clean aluminum foil.

### 7.4.3 Installing a New Electron Multiplier

*Note:* This is best to do when you also need to install a clean ion source.

Clean white gloves must be worn at all times. Never touch the electron multiplier or analyzer panel with bare hands. Using needle-nose pliers, lift up on the wire holding in the old electron multiplier and remove it from its lead. Install a new multiplier, making sure the lead is snugly attached. Place the pin back to hold it in place, and make sure your connections are tight. Tighten all screws, as needed.



Figure 42. Image of the HED control cable (yellow), the signal cable (black), and the HED power cable (white with orange tips). Disconnect all three before removing the analyzer.

### 7.4.4 Installing the MS Analyzer

Before installing the analyzer and closing the MS, a few things should be inspected. Check the MS for scorch marks indicating a short in the system. Run a gloved finger along the black ring-seal, pushing in any bumps. Use a flashlight and check the seal for any cracking or chipping—a damaged seal needs to be replaced. After a successful inspection, the analyzer panel can be installed. The analyzer panel is a little tricky to put back on; you need to put the top peg at the back of the panel into the opening before placing the bottom peg in its hole. Push the panel back into place, then tighten the two screws that hold it. Reattach the two electrical leads. The MS can now be pumped down. Proceed to [Section 7.2](#) before using the MS.

### 7.5 Adjusting the Radio Frequency

It might be necessary to adjust the radio frequency (RF) after installing a clean source and/or new electron multiplier. Adjust the RF values after allowing the MS to pump down ([Section 7.2](#)) for at least a day following the maintenance. Click **Tune and Vacuum Control**. Click **Execute**, then select **Optimize Quadrupole**. Choose **100 AMU** first, then **700 AMU** second.<sup>1</sup> The values for the low end (100 AMU) should be approximately 70–75. The values for the high end (700 AMU) should be approximately 500–510. If the values for 100 AMU and 700 AMU are low or high, obtain the target AMU by using a flathead screw driver to adjust the black screws just inside of the MS door near the source (Figure 43).

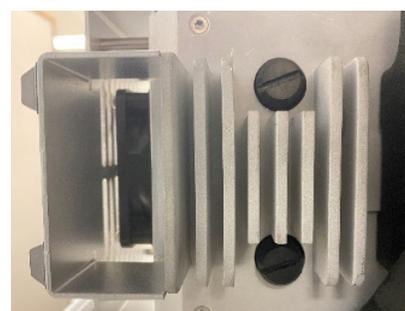


Figure 43. Use these black screws to adjust the radio frequency.

<sup>1</sup>AMU = atomic mass unit.

## 7.6 Changing the Rough Pump Oil

Vent the MS ([Section 7.1](#)). The rough pump oil should be drained and replaced at least once a year. It is advised to check the oil level ([Figure 44](#)) every week to catch potential leaks or problems with the pump. If the level is low or the oil looks dark, it is a good idea to change the oil even if it hasn't been a year since the last oil change.

Start by disconnecting the main vacuum hose. Remove the O-ring fitting and place it on a clean paper towel so it doesn't get dirty. Put on clean gloves and lift the pump up onto the end of a table, drain hole to the end of the table. Remove the filter cap with a pair of vise grips. Remove the blue plastic filter and replace it with a new one. Using the oil waste jug and oil funnel found under the sink, unscrew the black drain plug and drain the pump. Once it is completely drained, screw the drain plug back in tight. Using a clean funnel, slowly pour new Inland 45 oil (kept under the sink) up to the max line, making sure not to overfill it. Place the cap back on and tighten. The pump, along with the area you were working in, should be wiped down with some isopropanol, to ensure that there is no residual oil left behind. Open up the ballast and leave it loose for a few minutes to expel air. Then, place the O-ring back on the top of the port that the hose connects to. Now you can place the pump back on the tray and back in the spot where it operates. Place the hose on flat and evenly, and place the grippers back on and tighten them. Pump down the system ([Section 7.2](#)). Record the changing of the rough pump oil in the system's logbook, highlight it, and update the white board.

The used oil that was collected in the waste jug should be taken down to the oil waste container on the first floor of the East Building and emptied. The secondary oil waste jug, kept in **Room 317E**, should never be left with oil inside.

## 7.7 Refilling the Calibration Gas

There should be enough gas in the bulb to make the liquid touch the glass capillary tube on the side ([Figure 23](#)). Vent the MS ([Section 7.1](#)). After the system is vented, use needle-nose pliers to loosen the metal fitting. Get the perflurotributylamine that is stored in the refrigerator door. Using a pipet, add a few drops until it touches the insert (when held on its side). Put the fitting back on and tighten it using the needle-nose pliers. (Don't overtighten.) Record the refilling of the calibration gas in the system's logbook and highlight it.

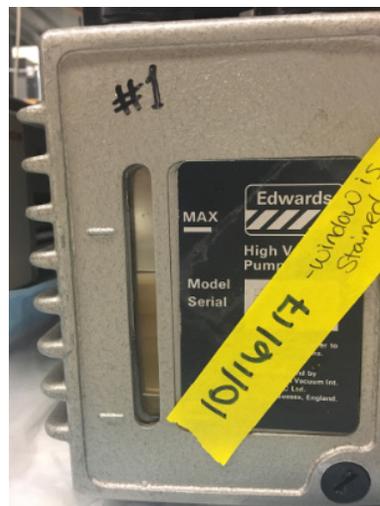


Figure 44. The oil level window on a GC/MS rough pump.

## 8 Troubleshooting

When it comes to troubleshooting problems with the GC/MS, it is best to go back to the most-recent thing you fixed and/or altered. Although most problems that will occur are not straightforward, there are signs and signals that help point the technician in the right direction.

### 8.1 Leaks

A leak in the system will show up in the air/water spectrum ([Section 4.2](#)) as high responses for all three masses (over 5,000 counts). It could also present itself as low responses in the standard run for a performance check. Cool the oven to 30°C, then open the GC. Use the ¼" wrench to loosen, then tighten, the MS interface, and to tighten the inlet. Check the air/water spectrum again. If the leak is still present, the union will have to be disassembled so the columns can be checked. A sheared column will have to be trimmed and will require a new ferrule. It is also advisable to check the head of the guard column in the inlet to make sure that the nut was not overtightened, causing the head of the column to shear off. A high tune background indicates activity in the system and/or possibly MS contamination. System activity means something in the columns is causing an unwanted reaction. The solution is to run a series of conditioning and standard runs to flush the columns out. Instead of running just standards, use a series of 6–8 conditioning runs, alternating with reproducibility standards (all in separate vials). Then check the tunes to see if the background has decreased. If not, try running another series of conditioning runs.

Leaks can also be detected and located using a can of Dust-Off (the white cans in the cabinet in the GC/MS lab), which contains difluoroethane (mass 47). In the MS control window, click **View**, then select **Tune/Vacuum Control**. Click **Manual Tune**, then select **More Parameters**. Select the three masses you want to monitor (47, 28, 32), and then click **Profile**. Spray small puffs of Dust-Off around the vacuum flange. Make sure to hold the can upright, and *do not shake the can* before spraying the flange. If you can see a big peak arise, you know that you don't have a good seal. You can also use this method around the interface nut, the vent valve, and the outside of the interface to see if a leak is there. If you detect a leak, vent the system, reseal the area if possible, and pump the system down. Then attempt the leak test again.

### 8.2 Signs That it is Time to Replace the Analytical Column

Although the cause is not always clear when you are troubleshooting problems, there are some clues along the way that help point you in the right direction. Column bleed, sensitivity, peak shape, and how noisy the baseline is can tell you a lot about the quality of the analytical column that is currently installed.

## 8.2.1 Checking Column Bleed

If you suspect the chromatography of your analytical column is becoming compromised, run an **AHP7A** standard on the **AHscan** method (using **aTune.u**, *not* **AHtune.u**) to look at the maximum column bleed. When the standard is finished running, enlarge the high end of the chromatogram's baseline and note its abundance. An abundance <100,000 is usually fine, but once it exceeds 150,000–200,000 you should consider replacing the column. High column bleed can affect not only the chromatography of the system, but can prematurely foul the MS ion source, causing other types of reduced performance.

## 8.2.2 Peak Shape

See [Section 5.3](#). If the peak shape is not improved after routine maintenance ([Section 2](#)), you should consider replacing the column.

## 8.2.3 Sensitivity

See [Section 5.3](#). If the sensitivity is not improved after routine maintenance ([Section 2](#)), you should consider replacing the column.

## 8.2.4 Noisy Baseline

Conditioning the column overnight should resolve baseline noise and help to achieve acceptable signal-to-noise ratios for the compound peaks anticipated in the analysis. In some instances, multiple days of conditioning may be needed. If the baseline is not improved after conditioning the column, you should consider replacing the column.

## 8.3 Signs of a Dirty Ion Source

A dirty ion source will cause peaks to tail, high-end response to drop, and resolution to deteriorate—even after column maintenance. This will be a trend, starting as a minimal problem and progressing until data are no longer be usable. Look at the resolution of double-peaked compounds, such as indeno(c123)pyrene in **AHP4A**. Good resolution will mean a deep valley between the two peaks. A dirty ion source will reveal itself by creating a shallower valley in this compound. Also look for persistent tailing, especially in the high-end peaks. If these are present, install a clean ion source ([Section 7.3](#)).

## 8.4 Checking/Replacing the Syringe

The syringe in the auto-sampler injector tower (Figure 45) may become defective, causing poor or no injection of samples/standards. The plunger may become sticky, and the needle or plunger will occasionally bend or break. To remove a defective syringe, unscrew the plunger nut and slide the plunger action up. Move aside the black lever holding the syringe in place, then carefully remove the syringe. Take care not to let any parts fall into the injector tower.

Before (re)installing a new syringe (Figure 46) in the auto-sampler, always make sure it is working properly by gently manually drawing up and expelling some solvent from one of the rinse vials 2–3 times. Once the syringe has been assembled and tested, insert the syringe needle into the guide at the bottom of the injector tower, then nestle the syringe barrel into place with the barrel's shoulder atop the green bracket. Move the black lever over the barrel. Finally, fully unscrew the nut in the plunger action, slide the action down over the syringe plunger, and tighten the nut securely. Occasionally the red **Fault** light on the injector tower will remain lit after the syringe is replaced. Unplug the power cord for the injector on the back of the GC for a few seconds, then plug it back in and the warning light should reset.



Figure 46. Replacement syringe.



Figure 45. Inside of the auto-sampler injector tower. The syringe is held in place by the plunger nut and black lever.

## 9 Helium

The GC/MS systems use Helium grade 5.0, “Ultra High Purity Grade,” as a carrier gas.

### 9.1 Helium Tanks

The helium tanks are stored in the gas cylinder bay on the second floor of the East Building, in the hallway of the north loading dock. Each system has two open tanks attached to a switching valve. If the pressure drops below approximately 200 pounds, the switching valve will kick on and switch tanks. The tanks should be manually switched when the pressure gets to 500 psi. Manually switch tanks by turning the knob with an arrow on the front of it to point to the full tank (Figure 47). When one tank has been depleted, disconnect that tank and switch it out for a new one. It is important to fully open the new tank even if it isn't in use—this way, if something happens to the other tank, the switching valve will kick in and begin to use the new tank.

Check the helium tanks weekly in order to monitor the status of each tank. During this check, it is a good idea to keep inventory of the number of full, unused tanks you have for backup. Ideally there should be a backup tank for every system. The key to the gas cylinder bay is found on the peg board behind the door of **Lab 253E**.

### 9.2 Helium Filters

The Restek helium gas filters are located on the north wall of **Room 317E** (Figure 48). An oxygen trap is on the back of each GC. These filters should be monitored frequently and replaced when their indicator colors change. The moisture indicator color is yellow/orange; it will change to clear when it becomes saturated. The oxygen indicator color is green; it will begin to turn gray when it becomes saturated.



Figure 47. (top) Helium gas tank bay for the GC/MS instruments. (bottom) Two open helium tanks hooked up to a switching valve.



Figure 48. Helium gas filters for all four lines supplying gas to the GC/MSes.

## 10 Purchasing/Inventory

An inventory of all consumables used to operate and maintain the GC/MSes should be conducted at least once a month. Backup supplies should always be kept on hand in case of emergency. The only item that cannot be kept as backup is the electron multipliers—due to their short shelf lives, they should only be ordered when needed.

## 11 Sample Storage/Archival

After the data have been reported, the sample sets can be removed from the refrigerator in **Room 317E** and brought downstairs to be archived in **ECP freezer #19** in **Lab 253E**. Locate the appropriate vial case in **ECP 19** for the set to be archived in. There are cases for **LCREP**, **PSEMP**, **Marine Mammal**, **Project 90/100**, and **Other**. The set name should be written on tape on the lid of the case, over the section where the vials will be kept. Make sure that the caps are on securely before placing them in the case. If there are not enough vials to fill the section, use paper towel to fill the void in order to hold the vials upright.



Acting U.S. Secretary of Commerce  
Wynn Coggins

Acting Under Secretary of Commerce  
for Oceans and Atmosphere  
Benjamin Friedman

Acting Assistant Administrator for  
Fisheries  
Dr. Paul N. Doremus

**February 2021**

[fisheries.noaa.gov](http://fisheries.noaa.gov)

OFFICIAL BUSINESS

National Marine  
Fisheries Service  
Northwest Fisheries Science Center  
2725 Montlake Boulevard East  
Seattle, Washington 98112